BACTERIOLOGICAL DIAGNOSIS

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TRANSLATED BY

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Bacteriological Diagnosis:

TABULAR AIDS FOR USE IN PRACTICAL WORK.

BY

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Vienna.

TRANSLATED AND AUGMENTED, WITH THE PERMISSION OF THE AUTHOR,
FROM THE SECOND GERMAN EDITION,

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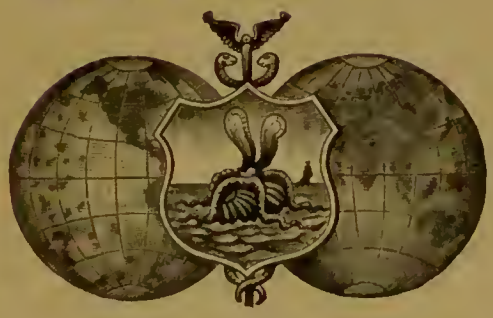

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TO

MY PROFOUNDLY ESTEEMED TEACHER,

DR. ROBERT KOCH,

THIS WORK IS RESPECTFULLY
DEDICATED.

PREFACE.

"The further bacteriological investigation advances, the more obvious becomes the fact that it is absolutely unallowable to base our diagnosis of a given bacteria upon anything short of a careful consideration of *all* its characteristics and properties, and especially when such a bacteria resembles another in one or more respects. Thus, there exist many forms of bacilli which, morphologically, are almost indistinguishable, but which, in pure culture on potato or gelatin, or on inspissated blood-serum, differ essentially one from another."

It is in daily practical work that one learns the truth of these remarks of Dr. Koch. And thus a well-defined demand has arisen for a work in which the specific differences of bacteriæ shall be tabulated for ready reference, resembling, in this regard, the "Chemical Analysis Chart" which the chemist finds so essential in his laboratory work. With this end in view, I have endeavored to arrange the following series of tables in as concise a form as possible, that the worker may at once inform himself as to the identity and significance of a given micro-organism.

There are blank forms occasionally interspersed, which may be filled out as further advance takes place.

Of the organisms that have been described, only those which possess marked biological, anatomical, or physiological points of differentiation are here considered. In the anatomical description and arrangement I have followed the method of Koch, which, doubtless, is the most trustworthy. First, the cover-glass preparation is studied; then the action in the moist chamber, by which procedure motility is ascertained; after which plate cultures are made, by means of which the next steps—pure tube cultures—are rendered possible. For the latter purpose 10-per-cent. meat-peptone gelatin and the 20-per-cent. meat-peptone agar-agar were used. The growth on potatoes and blood-serum has also been observed and noted, and occasionally that in bouillon.

In presenting the physiological attributes, only the most important have been selected. Anaerobiosis has been studied in the following manner: The contents of a tube of sterilized gelatin is poured onto a glass plate and allowed to harden, after which it is inoculated in long strokes with the germ under observation. Half of the plate is covered with sterilized mica, and in the course of a few days we get a characteristic picture. Some organisms will grow beneath the mica, and others not, thus proving them anaerobic or aerobic. Spore formation was studied on agar-agar in shallow, double watch-glasses, and on potatoes.

In selecting a classification I encountered some difficulties, and at last decided to adopt one that is quite arbitrary. But even this has not been strictly adhered to. In the first place, I made a division into the non-pathogenic and pathogenic, the former of which were again subdivided into those that did and did not liquefy gelatin. The pathogenic were subdivided into those that were and were not cultivated outside the animal body. As far as possible, an alphabetical arrangement has also been preserved.

Finally, as an appendix, I have a summary of the most generally distributed fungi, κατ' ἐξοχήν, by which term is meant all those micro-organisms which possess the true "twig" formation, differing in this regard from all bacteries, while the absence of chlorophyl is common to both. Their sequence here is controlled by the difference in their organs of fructification, which divide them into several species, from the more complicated forms to the simple yeast-cell, which has no mycel whatever. I will mention the actinomyces as a transition form—a bridge between the fungi and those pathogenic bacteries which, by living and multiplying within the human organism, play the chief factor in disease, possessing, as this organism does, the characteristics of the former and the pathogenic attributes of the latter. From among a hundred or more species of fungi only a comparatively few have been selected, but these are the ones most frequently encountered and possess the most pronounced characteristics.

The arrangement of these tables was commenced in company with Dr. F. G. Gade, of Christiania, who has given especial attention to the works of Rosenbach and Passet, and he has personally

gone over the ground with each respective organism. In all sincerity, I extend to him my most heart-felt thanks for his generous assistance, only regretting that we could not complete the work together.

Finally, I take especial delight in acknowledging the kind attentions of the Honorable Privicouncillor, Professor Koch, in whose laboratories these experiments were made, and to whose practical assistance and advice I shall ever feel indebted.

BERLIN, October, 1885.

PREFACE TO THE SECOND EDITION.

A LITTLE over a year has elapsed since the appearance of the first edition of these tablets, and, since that was exhausted in so short a time, I believe that I may safely accept my publisher's suggestion to send forth a second and revised edition. I can only trust that this will meet with as fair a reception as the previous edition, and that it may be found that I have profited by the advice and hints of my *critiques*. Special consideration was given to the observations of Professor Baumgarten and those of the late Professor Friedländer, and, in consequence, a different typographical arrangement has been selected, whereby space has been saved without the sacrifice of matter.

It may be thought by some that the number of micro-organisms might be extended. To this I will only say that only the most thoroughly studied and more-important species have been admitted. I have endeavored to preserve the original purpose of the book, namely, that of a guide to the student and an aid and a summary to the more-advanced worker.

One hundred and thirty-eight micro-organisms are considered. I may add here that contemporaneous literature has been thoroughly reviewed and applied in my own experiments. I have gained much from Flügge's vast work, "Micro-organisms," and, in many instances, have taken advantage of his admirable terminology.

The greatest difficulty encountered in this edition, as in the former, was the classification, and I have had to retain the same arbitrary one. I hope that the alphabetical index will increase the practical working of the following pages.

WIEN, June, 1887.

TRANSLATOR'S PREFACE.

THAT the work is used in Professor Cohn's laboratory and by other teachers, and that it met with an almost universal indorsement from the German medical press, is sufficient guarantee of its worth. The task of translation was undertaken from a conviction that the book would be useful to the American worker in the important and everdeveloping field of bacteriology, and with a hope that it might increase the number so engaged—"a consummation devoutly to be wished."

There are richer and more wondrous truths lying hidden in this direction than have as yet been dreamed of.

The arrangement of the text has been somewhat changed from the original, whereby a greater uniformity has been preserved. I have been tempted to admit several other recently-reported species to these pages, but have decided to allow further investigation to corroborate them. The few that have been admitted were added to the Italian translation by Titto Ferretti, and may be considered as fairly established.

Finally, I cannot refrain from acknowledging the great aid I have experienced from that most excellent work, the "National Medical Dictionary," and I most modestly compliment the eminent gentlemen upon the masterly manner in which they have acquitted themselves as editor and collaborators.

NORVAL H. PIERCE.

CHICAGO, 1890.

CONTENTS.

I. Non-Pathogenic Bacteria.

A. Liquefying Gelatin.

I. Non-Pathogenic Bacteria.

B. Not Liquefying Gelatin.

II. Pathogenic Bacteria.

A. Cultivated Outside the Animal Body.

II. Pathogenic Bacteria.

B. Not Cultivated Outside the Animal Body.

III. Fungi.

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I.

NON-PATHOGENIC BACTERIA.

A. Liquefying Gelatin.

1. Bacillus prodigiosus.

Place found.	In amyloid substances, moist bread, cooked potatoes, etc. Most probably it settles from the air onto the nourishing material; but, up to the present, they have not been demonstrated in the air.
Form and arrangement.	Very short bacilli, closely related to the coccus form; often considered as diplococci.
Motility.	Non-motile.
Growth.	
Gelatin.	*On Plates.*—Circular, granular colonies with smooth rims. Later, a red color is developed in the centre of the colonies. *Tube Culture.*—Rapid growth along the inoculation stroke. On the surface a red coloring matter is formed, which settles to the bottom when shaken. The growth liquefies the gelatin.
Agar-agar.	*Tube Culture.*—In the form of a pretty, purplish-red coating. The coloring matter does not enter the agar-agar.
Potatoes.	An especially luxuriant growth occurs in twelve hours at common temperature. The coloring is purple-red, which changes to a pretty fuchsin or greenish color endowed with a metallic lustre. Later, an odor of trimethylamine arises.
Blood-serum.	The same as on agar-agar, with the exception that it liquefies serum.
Temperature.	Thrives best at 25° C. When the temperature is raised the growth is decreased.
Rapidity of growth.	Grows very rapidly.
Spore formation.	Up to the present time no lasting spores have been found. Yet it is noteworthy that it retains the power of development for a long time after drying.
Aerobiosis.	Pigment formation is dependent upon the admission of air. Does not grow under mica plate.
Gas production.	
Gelatin reaction.	Very rapid liquefication.
Color production.	The pigment is red. When touched with acetic acid it becomes brighter and assumes the color of the bacillus indicus, which, treated with ammonia, returns to its original color.

2. Bacillus indicus. *Koch.*

Place found.	First found in India in the contents of a monkey's stomach.
Form and arrangement.	Very fine and short bacilli with rounded ends.
Motility.	Motile.
Growth.	
Gelatin.	*On Plates.*—Round, quickly-liquefying colonies, similar to No. 1. *Tube Culture.*—The growth liquefies the gelatin, which is colored a brick-red, and rests on the solid media.
Agar-agar.	*Tube Culture.*—In the form of a brick-red coating, which often possesses a whitish fimbria.
Potatoes.	After twelve to twenty four hours brick-red growth occurs, which is limited to the inoculation stroke.
Blood-serum.	Liquefies the serum, which is occasionally colored by the characteristic coloring matter.
Temperature.	Thrives best at 35° C.
Rapidity of growth.	Very rapid.
Spore formation.	
Aerobiosis.	Pigmentation depends upon admission of air. Does not grow under mica plate
Gas production.	
Gelatin reaction.	Very rapid liquefication.
Color formation.	Forms a brick-red coloring matter. When moistened with ammonia it assumes the color of the bacillus prodigiosus, which, treated with the acetic acid, returns to the original color.
Pathogenesis.	When injected in large amounts into the blood of rabbits, diarrhœa occurs a short time afterward, and death results in from three to twenty hours. At the section signs of severe gastro-enteritis are found.

3. Sarcina alba.

Place found.	Air and water.
Form and arrangement.	Small cocci, arranged in pairs and fours.
Motility.	
Growth.	
Gelatin.	*On Plates.*—Growth takes place slowly. The colonies are small, round, and white. *Tube Culture.*—A delicate growth along the inoculation puncture, which is surmounted on the surface by a whitish button-like formation.
Potatoes.	Grows very slowly in the form of a yellowish-white coating confined to the inoculation stroke.
Blood-serum.	
Temperature.	
Rapidity of growth.	Grows very slowly.
Spore formation.	
Aerobiosis.	Does not grow under mica plate.
Gas production.	
Gelatin reaction.	Liquefaction is only slight and takes place slowly.
Color production.	
Pathogenesis.	

4. Sarcina aurantiaca.

Place found.	Air.
Form and arrangement.	Small, hemispherical cocci, arranged in twos and fours, forming packets.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Small colonies. With low-power magnification a punctated appearance is observed. Colonies are round, with smooth edges. *Tube Culture.*—Liquefying slowly along the whole length of the inoculation stroke produces an orange-yellow color on the surface.
Potatoes.	Grows very slowly, of a yellow color, limited to the point of inoculation.
Temperature.	Does not thrive (or very imperfectly) at incubator temperature.
Rapidity of growth.	Grows very slowly.
Formation of spores.	
Aerobiosis.	Does not grow under mica plate.
Gas production.	
Gelatin reaction.	Liquefying.
Color production.	Forms golden-yellow coloring matter.
Pathogenesis.	

5. Sarcina lutea. *Schröter.*

Place found.	Air. In conjunctival sac.
Form and arrangement.	Very large cocci, arranged together in twos or fours. (Like bales of cotton.)
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Grows very slowly in small, round, yellowish colonies. *Tube Culture.*—Not liquefying. Growth very slow, of a yellowish color along the stroke, and slightly spreading laterally.
Agar-agar.	*Tube Culture.*—Not liquefying. Growth of a pretty lemon color
Potatoes.	Grows very slowly in sulphur-yellow colonies at the point of inoculation.
Temperature.	Thrives at temperature of blood.
Rapidity of growth.	Grows very slowly.
Spore formation.	
Aerobiosis.	Does not grow under mica plate.
Gas production.	
Gelatin reaction.	Grows slowly and liquefies very slightly.
Color production.	Forms lemon-yellow coloring matter.
Pathogenesis.	

6. Bacillus violaceus.

Place found.	Water.
Form and arrangement.	Slim staves with curved ends, the length of which is four times their width. They also form threads by continuity of individuals.
Motility.	Quite motile.
Growth.	
Gelatin.	*Plate Culture.*—Round colonies with smooth edges, in the middle of which is found the coloring matter. They liquefy gelatin. *Tube Culture.*—Liquefies in the form of a funnel, at the cusp of which is a violet granular mass.
Agar-agar.	*Tube Culture.*—Growth forms a gorgeous mass of a very pretty deep-violet color, spreading very quickly on the surface.
Potatoes.	Growth very slow, of a dark-violet, almost black color, confined strictly to the inoculation point.
Blood-serum.	Liquefies, forming violet coloring matter.
Temperature.	Does not thrive at a high temperature.
Rapidity of growth.	Grows slowly.
Formation of spores.	Spores of medium size.
Aerobiosis.	Same as No. 1.
Gas production.	
Gelatin reaction.	Liquefies.
Color production.	Forms dark-violet coloring matter.
Pathogenesis.	

6*. Bacillus coeruleus. *Allen Smith.* *Medical News, Philadelphia. December 31, 1887.*

Place found.	In the waters of the Schuylkill River.
Form and arrangement.	Very small, straight staves, 0.002 to 0.005 mm. in length and 0.005 in breadth, sometimes curved. Occurring occasionally in chains resembling leptothrix.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Small, superficial colonies, forming a delicate film of light-blue color. Slight liquefaction. *Tube Culture.*—Development occurs along the inoculation puncture and on the surface. The deeper colonies are colorless; the ones situated more superficially have an azure color, owing to their transparency. Slight liquefaction.
Potatoes.	The development is characteristic. Colonies are cup-shaped with well-defined margins, which lie on the surface of the potato. At first the color is dark blue, turning to a black as the culture grows older.
Temperature.	
Rapidity of growth.	
Spore formation.	
Aerobiosis.	The color productions depends upon the presence of air.
Gas production.	
Gelatin reaction.	Limited liquefaction.
Aniline reaction.	Colors well with all the aniline stains. The best is methylene violet. *Color Production.*—The bacilli produce an intense azure, which is unaffected by water, alcohol, or dilute acids. The pigment of the bacillus pyoscyanus, treated with soda or potassium, gives a red color, and with ammonia a violet color. That of the bacillus violaceus is dissolved in alcohol. That of the micrococcus cyaneus penetrates the potato, and is dissolved in water, coloring it at first green, later azure.
Pathogenesis.	Non-pathogenic.

7. Bacillus ruber.

Place found.	Water.
Form and arrangement.	Medium-sized bacilli with rounded ends, often arranging themselves in long threads.
Motility.	Very motile.
Growth.	
Gelatin.	*Plate Culture.*—Round, fine, granular colonies, with smooth rims, colored in middle; liquefies. *Tube Culture.*—Grows slowly and forms a reddish-brown coloring; liquefies.
Agar-agar.	*Tube Culture.*—Growth of brown-red color, which spreads quickly over the entire surface.
Potatoes.	Growth of a pretty, reddish-brown color, *not* confined to inoculation point.
Blood-serum.	Liquefies, and produces a reddish-brown color.
Temperature.	Does not thrive at high temperature.
Rapidity of growth.	Grows very slowly.
Spore formation.	
Aerobiosis.	Does not grow under cover-glass.
Gas production.	
Gelatin reaction.	Liquefying.
Color production.	Forms reddish-brown coloring.
Pathogenesis.	

8. Green-yellow bacillus.

Place found.	Water.
Form and arrangement.	Small, fine staves.
Motility.	Very motile.
Growth.	
Gelatin.	*Plate Culture.*—Colonies round, white in the middle; forms funnel-shaped depressions on the media, which often assumes a greenish-yellow color for 4 to 5 millimetres around the colonies. *Tube Culture.*—Hardly visible along the inoculation stroke, where it grows very slowly; on the surface of media it grows quicker and forms bubbles. The firm gelatin around the colonies after awhile assumes a diffuse, greenish-yellow fluorescence.
Potatoes.	Growth of muddy-yellow color on inoculation stroke, the adjacent surface assuming a light-brown color.
Temperature.	Does not thrive at high temperature.
Rapidity of growth.	Grows quickly.
Spore formation.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Liquefying.
Color production.	Forms greenish-yellow fluorescent coloring matter.
Pathogenesis.	

9. Gas-forming bacillus.

Place found.	Water.
Form and arrangement.	Small staves.
Motility.	Very motile.
Growth.	
Gelatin.	*Plate Culture.*—Liquefies quickly. Colonies of medium size, forming shallow depressions; with low power they are seen to have grayish contents, and gas-bubbles are often visible. *Tube Culture.*—Growth occurs quickly, resembling somewhat the shape of a stocking. In the solid gelatin alongside the inoculation stroke gas-bubbles occur.
Potatoes.	
Blood-serum.	
Temperature.	Does not thrive at high temperature.
Rapidity of growth.	Grows very fast.
Spore formation.	
Aerobiosis.	
Gas production.	Forms gas-bubbles.
Gelatin reaction.	Liquefies.
Color production.	
Pathogenesis.	

10. Liquefying bacillus.

Place found.	Water.
Form and arrangement.	Short, rather thick staves, with rounded ends.
Motility.	Very motile.
Growth.	
Gelatin.	*Plate Culture.*—Round colonies, the edges of which are smooth; the centres are occupied by a slimy mass; liquefaction begins in excavations, which quickly spread. After a time an odor of putrefaction arises. *Tube Culture.*—Grows quickly along the entire inoculation puncture, funnel-formed, and whitish in color. Air-bubbles occur at the base of the growth.
Potatoes.	Growth colored a light yellow.
Blood-serum.	
Temperature.	Does not thrive at high temperature.
Rapidity of growth.	Grows very fast.
Formation of spores.	
Aerobiosis.	Does not grow under cover-glass.
Gas production.	
Gelatin reaction.	Liquefying.
Color production.	On potatoes a light yellow is produced.
Pathogenesis.	

II. Bacillus phosphorescens. *Fischer.* *Zeitschr. f. Hygiene, Bd. II, 54.*

Place found.	Sea-water.
Form and arrangement.	Small, thick staves, from two to three times as long as they are broad, with rounded ends, and having a tendency to form threads.
Motility.	Extremely motile.
Growth.	
Gelatin.	*On Plates.*—After thirty-six hours small, round, grayish-white dots are formed. Under a low power the colonies appear circular and sharply defined, and of a sea-green color, with a rosy shimmer. Further advanced, they become distinctly granular, of a dirty-yellow color, and the edges appear wavy and bent. *Tube Culture.*—On the fourth day a cup-shaped depression, about the size of a hemp-seed, is formed on the point of inoculation, which contains gas. Along the inoculation stroke a dim, whitish-gray, thread-like line appears. Later, the liquefaction advances, and, in old cultures, a thin, muddy-yellow scum floats on the surface.
Agar-agar.	*On Plates.*—As a grayish-white coating.
Potatoes.	Growth occurs at 15° to 20° C. as a broad, thin, white film.
Blood-serum.	The growth, which first appears as a grayish-white line, often increases to a deep, wavy stripe, reaching a breadth of ½ to 1 centimetre, and setting in a depression that it has caused on the serum. Cooked fish, meat, and blood are favorable nourishing media.
Temperature.	20° to 30° C. is the most favorable temperature. Under 10° C. the growth ceases, and at 55° C. it dies. They withstand a temperature of 15° C. for three hours.
Rapidity of growth.	Very slowly.
Spore formation.	Spore formation not as yet observed.
Aerobiosis.	Do not grow under mica plate nor in an atmosphere of carbonic acid.
Gas production.	
Gelatin reaction.	Liquefies.
Color production.	Common aniline colors are good for stains; in the centre uncolored spots remain, the same as in the bacilli of rabbit septicæmia.
Pathogenesis.	Do not possess pathogenic qualities. *Physiological.*—Animal matter in the presence of certain natural salts seems to form a good nourishing ground. The bacilli of old cultures emit a characteristic phosphorescence in the dark, which is dependent upon the presence of air, a certain degree of moisture, and a proper temperature (25° to 30° C. is best). Putrefaction and a temperature under 0° C. destroy the phenomena.

12. Proteus vulgaris. *Hauser. Ueber Fäulnis Bakterien. Leipzig. 1885.*

Place found.	In putrefying animal matter and in meconium (Escherich).
Form and arrangement.	Slightly-bent staves, 0.6 m. in breadth and of very variable lengths up to 3.75 m. They often form long, twisted threads, resembling braided hair. Frequently involution forms are noticed.
Motility.	Very motile and possess long cilia.
Growth.	
Gelatin.	*On Plates* (5 per cent.).—Colonies are yellowish-brown in color, From their circumference they throw off outshoots which resemble bunches of hair. Each colony rests in an area of liquefied gelatin. The intertwined processes gradually spread out until they occupy the entire surface of gelatin. (This arrangement is made visible by the microscope.) Peculiarly-shaped zooglœa forms are often seen in the gelatin. *Tube Culture.*—Liquefaction occurs evenly along the puncture. After some time the entire contents are liquefied. On the surface a grayish-white cloud appears, while the greater part of the culture sinks to the bottom as larger and smaller particles.
Agar-agar.	*Tube Culture.*—In the form of a fast-spreading, moist, lustrous, grayish-white, thin coating.
Blood-serum.	Growth forms a dirty, smeary layer.
Temperature.	Grows best at 20° to 24° C. It retains its power of development after being subjected to a temperature of 15° to 20° C. for fifty-five hours.
Rapidity of growth.	Grows very rapidly.
Spore formation.	Spore formation not observed, although the cultures retain their vitality after being dried in thin layers.
Aerobiosis.	Grows as well in hydrogen as in an atmosphere of carbonic acid although very slowly.
Gelatin reaction.	Liquefaction occurs very rapidly and is unlimited.
Pathogenesis.	Intra-venous and subcutaneous injections in rabbits and guinea-pigs produce toxic symptoms. *Physiological Attributes.*—On fresh as well as on boiled and sterilized meat the bacilli cause putrid decomposition.

13. Proteus mirabilis. *Hauser. Ueber Fäulnir Bakterien. Leipzig. 1885.*

Place found.	In putrefying animal substances.
Form and arrangement.	Staves 0.6 m. in breadth, but varying greatly in length, some are nearly round. Involution forms occur more frequently than in No. 12, and take the shape of spheres or pears, measuring from 3.75 to 7.0 m. in diameter.
Motility.	Motile.
Growth.	
Gelatin.	*Plate Culture.*—Occurs as a rounded, white coating, which, under low power, gives a brownish, granulated appearance. The periphery slopes to the gelatin in a series of gradations or steps, the last of which is marked by wavy inlets. From the edges the individuals swarm over the surface; these are less motile than in No. 12. Zoogloœa also occur. *Tube Culture.*—In the periphery of the inoculation stroke there arises concentric zones composed of bacilli and threads. After forty-eight hours the surface colonies amalgamate and form a compact, succulent, shining coating over the gelatin, which, after two or three days, becomes completely liquefied.
Potatoes.	
Blood-serum.	
Temperature.	As in No. 12.
Rapidity of growth.	Grows rapidly.
Formation of spores.	As in No. 12.
Aerobiosis.	As in No. 12.
Gas production.	
Gelatin reaction.	Liquefies, but more slowly than No. 12.
Color production.	
Pathogenesis.	As in No. 12.

14. Bacillus sublilis (hay bacillus). *Ehrenberg.*

Place found.	In hay infusion.
Form and arrangement.	Bacilli 5 to 20 m. long and a third as thick, resembling bacillus anthracis, but somewhat narrower, with rounded ends, and developing into long threads. They are provided with flagellæ.
Motility.	Motile.
Growth.	
Gelatin.	*Plate Culture.*—Round colonies, with a halo-like periphery, whitish in color, and liquefying the gelatin. *Tube Culture.*—Grows fast and liquefies all the gelatin.
Agar-agar.	*Tube Culture.*—Very characteristic. On account of the pressure exerted by the bacilli upon themselves, caused by their rapid multiplication in the resisting media, a stiff, easily-removable, wrinkled surface growth is formed, that may be compared to that of the bacillus of potatoes in appearance.
Potatoes.	Grows diffusely on the entire surface as a moist, white, cream-like veil. When the soil is impoverished spore formation occurs.
Blood-serum.	Liquefies the serum and forms a wrinkled membrane on the surface.
Temperature.	Is capable of developing between 10° to 45° C. Grows best at 30° C.
Rapidity of growth.	Grows fast.
Spore formation.	Spores 1.2 m. in length, 0.6 m. in breadth. Staves sprout in the direction of long axis of spores. (Prazmovsky.)
Aerobiosis.	Does not grow under mica plate. Has a strong affinity for oxygen.
Gas production.	
Gelatin reaction.	Liquefying.
Color production.	
Pathogenesis.	Possesses no pathogenic qualities. Spores introduced into circulation are soon removed, or conducted to the liver or spleen, where they remain without influence. (Wyssokowitsch.)

14*. Bacillus scarlatinæ. *Klein, Edington.* *Jour. des Connaisance Med., August 25, 1887.*

Place found.	Bacilli 2 to 5 m. long and 0.4 to 0.5 m. in thickness; isolated until third day. Mixed with cocci after four to five days.
Form and arrangement.	
Motility.	
Growth.	
	Plate Culture.—After twenty-four hours deeply-seated colonies appear, which send forth delicate processes toward the surface. The gelatin is liquefied from the centre to the periphery.
Gelatin.	*Tube Culture.*—After twenty-four to thirty-six hours the gelatin becomes turbid along the inoculation stroke, and a liquefaction occurs in the "funnel form." On the surface a tough, thick film, which is ribbed, and possesses spots of yellowish brown. The bacilli are mixed with spores. After about a month the superficial growth falls to the bottom and disappears.
Bouillon.	The same as on gelatin, except that the margin of the film is markedly elevated to the extent of nearly 1 cm.
Potatoes.	At 18° C., after twenty-four hours, the growth forms a yellowish-white coating. Later, the centre assumes a lemon color, which by degrees extends to the periphery. Afterward the centre becomes a yellowish ochre, and at last a coffee color. After forty-eight to sixty hours the growth consists almost exclusively of spores.
Blood-serum.	Development is slight, and not especially characteristic.
Milk.	Forms a pretty skin over the surface, with precipitation of the casein, which is later dissolved, disappears, and the media becomes clear.
Temperature.	Grows at an ordinary temperature.
Rapidity of growth.	Grows slowly at 16° C., rapidly at 19° C.
Spore formation.	Forms oval spores from 1 to 1.5 m. in length and 0.5 to 0.75 m. in width.
Aerobiosis.	
Gas production.	
Gelatin reaction.	Liquefies.
Aniline reaction.	Colors best with a solution of methylene blue.
Pathogenesis.	Experiments carried on by the ingestion, subcutaneous injection, and scarification of scarlatinal blood on calves were followed by negative results. A committee called to pass judgment upon it declared that the bacillus scarlatinæ is an organism absolutely different from any other now known, but that it is morphologically identical with the streptococcus scarlatinæ of Klein.

15. Root bacillus.

Place found.	In the top layer of garden- or field- earth.
Form and arrangement.	Short bacilli, with rounded ends, about thrice as long as wide; often form long threads and chains.
Motility.	Very slightly motile.
Growth.	
Gelatin.	*Plate Culture.*—After two days veil-like, not very definitely circumscribed, whitish colonies develop, which spread quickly. They resemble somewhat a mycel fungus. Under a low power the colonies are seen to consist of an intertwining net-work of threads. *Tube Culture.*—At first the growth throws out runners and threads around the inoculation puncture, so that the whole "looks like an inverted fir-tree;" later, the growth spreads on the surface of gelatin, and then quickly comes to replace the entire contents of the tube. In old cultures there is a thick skin over surface, under which is a clear layer. On broken ground they appear as whitish spots. (Fränkel.)
Potatoes. Blood-serum.	Grows very slowly in the form of whitish, greasy scum, confined to the inoculation point. Spore formation abundant.
Temperature.	Thrives at incubator temperature.
Rapidity of growth.	Grows fast.
Spore formation.	Spores medium in size.
Aerobiosis.	Does not grow under mica plate.
Gas production.	
Gelatin reaction.	Liquefying.
Color production.	
Pathogenesis.	Even in large masses it is not pathogenic.

16. Clostridium fœtidum. *Liborius.* *Zeitschr. f. Hygiene, Bd. II, 160.*

Place found.	In the œdema exudate of mice which had been vaccinated with garden-earth, old cheese, and cow-excrement.
Form and arrangement.	Bacilli, 1 m. in thickness and of variable length, occasionally growing in threads.
Motility.	Very motile.
Growth.	
	They are easily obtainable when anaerobic conditions exist.
Gelatin.	*Plate Culture.*—Forms round, irregularly-limited colonies, which at an early date liquefy the gelatin immediately in contact with the growth, after which they form spherical bodies which contain in their interior an opaque liquid.
	Tube Culture.—Small, yellowish-white colonies, sending out short threads and lying in irregular heaps.
Agar-agar.	At the lower portion of the inoculation puncture a homogeneous cloudiness occurs in the media, after which isolated, irregularly-branching colonies are to be observed.
Potatoes.	
Blood-serum.	
Temperature.	
Rapidity of growth.	Grows fast.
Formation of spores.	Bacilli formed from the growth of spores, which are oval, strongly refractive, and of larger diameter than the œdema bacillus.
Aerobiosis.	Exquisitely anaerobic, yet not so sensitive to oxygen as the œdema bacillus.
Gas production.	Produces always a gas, which is decidedly offensive.
Gelatin reaction.	Liquefying.
Color production.	
Pathogenesis.	

17. Potato bacillus.

Place found.	Potatoes.
Form and arrangement.	Small, thick rods, with rounded ends, often lying two together or forming chains of four. Growing on gelatin or agar-agar, the ends are colored more than the centres.
Motility.	Quite motile.
Growth.	
Gelatin.	*Plate Culture.*—Round colonies, which have in their centre a yellowish film, quickly spreading through the media, which it liquefies. *Tube Culture.*—On the surface it forms a scum. The gelatin is quickly liquefied, the liquefaction beginning at the point of inoculation.
Potatoes.	Quickly growing, at first as a moist layer, which afterward turns to a tough mass, lying in folds and wrinkles. The mass is found to consist of long threads intertwined. In this stage spore formation is abundant.
Blood-serum.	
Temperature.	
Rapidity of growth.	Grows fast.
Production of spores.	Forms spores which appear as shining, oblong bodies.
Aerobiosis.	Does not grow under mica plate.
Gas production.	
Gelatin reaction.	Liquefying.
Color production.	
Pathogenesis.	

18. Bacillus megaterium. *de Bary.* *Vgl. Morphologie u. Biolog. de Pilze.*

Place found.	On cooked cabbage-leaves.
Form and arrangement.	Slightly-curved staves, 2 to 5 m. in thickness and about four times as long; ends rounded; often form chains of ten. They possess a peculiarity that is always present, *i.e.*, a granulation of the cell-contents. They have a strong tendency to bring forth involution forms.
Motility.	Movements take place in a peculiar, creeping manner, reminding one of the amœba.
Growth.	
Gelatin.	*Plate Culture.*—Small, round, liquefying colonies. *Tube Culture.*—Rapid growth. Liquefaction does not begin along the whole length of the inoculation puncture, but in a funnel form, beginning at the top surface.
Agar-agar.	*Tube Culture.*—Grows as a whitish scum on the surface, coloring the agar-agar dark.
Potatoes.	Grows very fast in the form of yellowish-white, cheesy colonies, confined to the point of inoculation. Spore formation quite abundant.
Blood-serum.	
Temperature.	Grows best at 20° C.
Rapidity of growth.	Grows fast.
Formation of spores.	Increase by transverse division and polar spore formation.
Aerobiosis.	Does not grow under mica plate.
Gas production.	
Gelatin reaction.	Liquefying slowly.
Color production.	
Pathogenesis.	

19. Bacterium graveolens. *Bordoni-Uffreduzzi.*

Fortschritte der Med. 1886. S. 157.

Place found.	In the epidermis from between the toes.
Form and arrangement.	0.8 m. long and broad.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Growth occurs at room-temperature in the form of irregular, whitish-gray colonies, which quickly liquefy the gelatin and give forth a disgusting odor similar to that which arises from the feet. Later, they assume a greenish-yellow color.
Agar-agar.	*Plate Culture.*—The same as on gelatin.
Potatoes.	Forms grayish, very foul colonies.
Blood-serum.	Liquefying. Growth the same.
Temperature.	
Rapidity of growth.	Growth very rapid.
Spore formation.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Liquefying.
Color production.	A greenish-yellow color develops late.
Pathogenesis.	

20. Bacillus butyricus (butyric-acid bacillus). *Hüppe.*

Mittheilg. a. d. Kais. Ges.-Amt. Bd. II.

Place found.	Fleshy roots, old cheese.
Form and arrangement.	Staves of varied lengths, somewhat bent, growing in threads.
Motility.	Very motile.
Growth.	
	Plate Culture.—In the deeper portions, pretty, yellowish heaps, which amalgamate into a seedy, brown mass with rounded corners. By their energetic action in liquefying the gelatin the observation of individual colonies is soon rendered impossible.
Gelatin.	*Tube Culture.*—Growth elaborate, yellow, and quickly liquefying. The growth spreads from the entire inoculation point. On the surface of the gelatin the growth assumes the form of a thin, whitish-gray skin, which lies in delicate folds. The chief growth occurs, however, in the lower liquefying layers.
Agar-agar.	*Plate Culture.*—Growth forms a yellowish, smeared coating.
Blood-serum.	
Temperature.	Grows most luxuriantly between 35° to 40° C., less so at 30° C.
Rapidity of growth.	Grows very rapidly.
Spore formation.	Occurs most abundantly at 35° to 40° C. The spore formation occurs at the end and at right angles with the long axis of the bacillus.
Aerobiosis.	
Gas production.	
Gelatin reaction.	Liquefying.
Color production.	
Pathogenesis.	*Physiological Attributes.*—Brings the casein of milk to the curdled condition of rennet, dissolves the albuminous body, and transforms it into a peptone, together with several bi-products, among them being ammonia,—the cause of the bitterness of sour milk. They also are the cause of butyric fermentation, and they are the active agents in the fermentation of sauer-kraut.

21. Clostridium polymyxa, Clostridium butyricum. ***Prazmowsky, Pasteur.*** *Comp. rend. 1861–1879. Prazmowsky. Untersuch. über die Entwickelungsgeschichte und Fermentwirk. ein. Bak. Leipzig. 1880.*

Place found.	In putrefying plant infusions. In fossils of the carbonaceous period. (Van Tieghem.)
Form and arrangement.	Large, thick staves, with rounded ends, from 3 to 10 m. in length and 1 m. in breadth, frequently arranged in chains.
Motility.	Very motile.
Growth.	
Gelatin.	Attempts at experimental cultivation on solid media not as yet thoroughly successful.
Potatoes.	
Blood-serum.	
Temperature.	Five minutes at boiling-point is sufficient to kill the spores.
Rapidity of growth.	
Spore formation.	At the stage of spore formation the bacilli assume the shape of spindles. At the moment of germination the limiting membrane bursts, which membrane is often dragged about by the young staves.
Aerobiosis.	Exquisitively anaerobic.
Gas production.	Those gases issuing from the gelatin and agar-agar tubes are of an unpleasant odor, resembling butyric acid.
Gelatin reaction.	
Color production.	Possess a reaction similar to granulose, *i.e.*, when in contact with aqueous solution of iodine are colored from a deep indigo blue to a dark violet.
Pathogenesis.	*Physiological Attributes.*—In solutions of starch, sugar, dextrin, or salts of lactic acid it produces an abundance of butyric acid, carbonic acid, and water. According to Fitz, the bacilli are also able to dissolve casein.

22. Miller's bacillus. *D. Med. Wochenschr. 1884. Nos. 36 and 48.*

Place found.	From carious dentine, besides four other fungi (α, β, γ, δ) already cultivated.
Form and arrangement.	Delicate, more or less bent staves. Lying two or four together, forming **S** figures occasionally, more frequently **O** shapes, also broken and unbroken spiral threads.
Motility.	Non-motile.
Growth.	
Gelatin.	*Plate Culture.*—Growth does not occur on surface. Liquefying. Does not cause the gelatin to melt away or evaporate. *Tube Culture.*—Growth similar to that of Finkler-Prior's bacillus.
Potatoes.	Not especially characteristic.
Blood-serum.	
Temperature.	
Rapidity of growth.	
Spore formation.	
Aerobiosis.	
Gas production.	Produces, together with the four associate fungi (α, β, γ, δ), an artificial caries that is not to be distinguished from the natural.
Gelatin reaction.	Liquefying.
Color formation.	
Pathogenesis.	

23. Micrococcus aerogenes. *Miller.* *Deutsche Med. Wochensch. 1886. No. 8.*

Place found.	From the digestive tract.
Form and arrangement.	Large oval cocci.
Motility.	Non-motile.
Growth.	
Gelatin.	*Plate Culture.*—Most frequently form round colonies of a dark color, smooth in contour, with irregularities like miniature inlets at their circumference. The characteristics which distinguish their growth are the leopard-like spots occurring in the colonies. *Tube Culture.*—Growth occurs along the entire inoculation puncture, of a brownish-yellow color, forming on the surface a button-like growth of a whitish color. After some time the gelatin is slowly liquefied.
Agar-agar.	*Tube Culture.*—Forms a yellowish-white, pulp-like surface growth.
Potatoes.	Grows over the surface as a yellowish-white, pulp-like mass, with irregular edges.
Blood-serum.	
Temperature.	
Rapidity of growth.	Grows rapidly.
Spore formation.	
Aerobiosis.	Grows with air admission slightly restricted.
Gas production.	Forms gas in carbohydrates.
Gelatin reaction.	Slightly liquefying.
Color production.	
Pathogenesis.	*Physiological Attributes.*—They possess a marked resistance to acids, so that they may be exposed for hours to an artificial digestive juice without impairing their development.

24. Streptococcus coli gracilis. *Escherich.* *Die Darmbakterien des Sauglings und ihre Beziehung zur Physiol. der Verdauung. Stuttgart. 1886.*

Place found.	In the alimentary canal and fæces of the carnivora; also in the meconium of the same after it has been exposed to infection from the air.
Form and arrangement.	Cocci, 0.2 to 0.4 m. in diameter, arranged in long S-shaped chains, consisting of from 6 to 20 members.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Small, dark colonies with sharp contour, at first round, later irregular, tunneling the gelatin in course of liquefaction. *Tube Culture.*—Liquefies along inoculation point in tubular form. In from six to eight days there appears at the bottom of the completely liquefied gelatin a precipitate of fine, whitish, seed-like particles.
Agar-agar.	*Tube Culture.*—Growth scant, occurs on the surface.
Potatoes.	Very slight and soon ceases. Growth occurs in the form of small, whitish buttons.
Blood-serum.	Slight growth in the form of small scales.
Temperature.	Grows best at incubator temperature.
Rapidity of growth.	Grows tolerably fast.
Formation of spores.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Very rapidly liquefying.
Color production.	
Pathogenesis.	

I.

NON-PATHOGENIC BACTERIA.

B. Not Liquefying Gelatin.

25. Micrococcus candicans. *Flügge.* *Mikroorganismen. Leipzig. 1886. S. 173.*

Place found.	Air. Extremely frequent accidental contamination of plate cultures.
Form and arrangement.	Very large, round cocci, occurring in irregular heaps.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Colonies form after two days below the surface of inoculation stroke of a yellowish-white color, 0.4 to 0.5 m. in diameter; on the surface they are of a pure, milk-white color, smooth, resembling a drop of lac. Under a low power they appear irregular and finely granular, lighter at the periphery, and dark brown at the centre. Those occurring at a depth appear more regularly round, possess smooth edges, and are of a dark-brown color, with a scarcely-perceptible surface granulation. *Tube Culture.*—Forms along the inoculation puncture a confluent whitish mass, and at its entrance a button-like elevation (rail culture).
Potatoes.	
Blood-serum.	
Temperature.	
Rapidity of growth.	Grows fast.
Spore formation.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Color production.	
Pathogenesis.	

26. Micrococcus versicolor. *See No. 25.*

Place found.	Frequent in the air.
Form and arrangement.	Small cocci, occurring in twos or heaps.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—After twenty-four hours white dots occur, at the bottom of the stroke, changing in two days to yellow, bullet-like colonies as large as 1 mm. Under a low power these appear round, sharply defined, of a yellowish-brown color, opaque and granular. On the surface the colonies are from 2 to 6 mm. in size, and after four or five days these may attain the size of even 10 mm.; they occur in heaps, smooth and irregular in outline, sometimes square. When they occur as a coating over the surface, it has a smeary appearance, is shining, and of a yellowish-green color. The color varies, however, according to the degree of illumination, giving a greenish, bluish, or mother-of-pearl shimmer. *Tube Culture.*—Small, spherical colonies, yellowish in color; on the surface a shimmering coating, like mother-of-pearl, with irregular edges.
Potatoes.	
Blood-serum.	
Temperature.	
Rapidity of growth.	Grows fast.
Spore formation.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Color production.	Yellowish green, or mother-of-pearl.
Pathogenesis.	

27. Sarcina ventriculi. ***Goodsir (1842), Falkenhaim.*** *Archiv f. Exper. Path. u. Pharmok., Bd. XIX, S. 339.*

Place found.	In the stomach-contents of man and animals.
Form and arrangement.	Colorless or yellowish-brown, round or slightly-oval cells, having an average diameter of 25 m., united in groups of four or multiples thereof, producing cubes with round edges.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—After thirty-six to forty-eight hours, round, yellowish colonies occur. From these colonies we may have colorless, bullet-like cocci, diplococci, and tetrads, but no cubes or packets.
Hay infusion.	Forms a skum consisting of brownish scales, and a flaky precipitate, both containing, in contradistinction to all other media, distinct cubes and packets.
Potatoes.	After twenty-four hours growth occurs along inoculation stroke in small, round, colorless, dry colonies, which, later, become yellowish, but always confined to point of inoculation.
Blood-serum.	Small, round, slightly-prominent, pale-yellow colonies.
Temperature.	
Rapidity of growth.	Grows fast.
Formation of spores.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Color production.	The outer covering gives with iodine and sulphuric acid or with iodine-chlor.-zinc solution a reddish-violet color (cellulose reaction).
Pathogenesis.	

28. Micrococcus ureæ. *Pasteur, Van Tieghem.* *Jaksch, Zeitschr. f. Phys. Chemie, Bd. V, S. 395. Leube u. Grasser, Virchow's Archiv, Bd. C, S. 556.*

Place found.	From ammoniacal urine and from air.
Form and arrangement.	Cocci 0.8 to 1 m. in diameter, often in form of diplococci, and grouped together in tetrads; also frequently occur in chains, in the form of a rosary and in zooglea. (Von Jaksch.)
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—After twenty-four hours, white, mother-of-pearl-like spots occur, about the size of a millet-seed, with a smooth, shining surface, and sharply-defined boundary. At the temperature of room they develop, after ten days, to the size of a three-cent piece. They are slightly elevated above the surface of the gelatin, giving the impression of a drop of stearine having fallen onto it. *Tube Culture.*—Grows in the form of a thin, tough thread. In old cultures an insipid, paste-like odor is noticeable.
Potatoes.	They grow best in Jaksch's artificial nourishing fluid. Its composition is as follows: 1 litre of water; $\frac{1}{75}$ gm. $MgSO_4$; $\frac{1}{8}$ gm. acid phosphate of kalium; 5 gm. natrium-tartrate of kalium; 5 gm. urine.
Blood-serum.	
Temperature.	Thrives best at 30° to 33° C. At 0° C. no growth. Can be preserved at 15° C. for days without losing its power of reproduction. (Von Jaksch.)
Rapidity of growth.	Grows quickly.
Spore formation.	
Aerobiosis.	Requires acids for development. Growth more abundant on the surface than below.
Gas production.	
Gelatin reaction.	Non-liquefying.
Color production	
Pathogenesis.	*Physiological Attributes.*—Decomposes urine into carbonic acid and ammonia (carbonate of ammonia).

29. Bacteria ureæ. *See No. 28.*

Place found.	From urine undergoing ammoniacal decomposition.
Form and arrangement.	Thick staves with rounded ends, mostly 2 m. long and 1 m. thick.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—On the second day a small, nearly transparent spot appears, which, in ten days, spreads to the size of a dime. They give an appearance to the surface of the gelatin similar to that of glass upon which we have blown our breath. The growth spreads from the inoculation point in irregular circular zones, the last of which is escaloped. *Tube Culture.*—After ten days a very thin, gray-colored growth occurs along the inoculation puncture. The surface growth is seldom extensive. Older cultures have a peculiar odor resembling herring-brine.
Potatoes.	
Blood-serum.	
Temperature.	
Rapidity of growth.	Grows unusually slow.
Spore formation.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Color production.	
Pathogenesis.	*Physiological Attributes.*—Decomposes urine into carbonate of ammonia, and more energetically than No. 28.

29 *. Bacteria of thready urine (bacillo gliscrogene). *Malerba, Sanna-Salaris, Melle.* *Giornale Internazionale delle Scienze Med., Fasc. II. Napoli, 1888.* *Melle.* *Ref. Med., August 30, 1888.*

Place found.	In thready urine.
Form and arrangement.	Micrococci elongated to the form of bacteria; at the period of cessation of growth a diplococcus-form appears, 1.14 to 1.57 m. long and 0.11 m. in diameter.
Motility.	Motile.
Growth.	
Gelatin.	*Plate Culture.*—After two days punctiform round colonies arise, which enlarge slowly and are granular. Some contain gas-bubbles at their centres; others are biconvex. The growth becomes corolla-formed and may be seen with the naked eye. The colonies later become umbellate and enlarged by the addition of concentric layers, the margins of which are smooth. Innumerable bubbles are formed at the bottom, which gradually rise to the surface, displacing the gelatin and breaking the colonies. *Tube Culture.*—Nail culture made up of superimposed spheres. They are tenacious, glutinous, and difficult to detach.
Agar gelatin.	*Tube Culture.*—After three to five days, granular. Moniliform, opalescent colonies develop along the inoculation stroke. At 37° C., after twenty to twenty-four hours, a thready liquid is deposited at the bottom.
Agar gelatin peptonized.	*Tube Culture.*—After twenty to twenty-four hours development is apparent in the form of sessile tufts, accompanied by numerous bubbles. A whitish pellicle at length forms on the top of a filamentous liquid.
Bouillon.	After twenty-four hours a diffused turbidity appears and the bouillon becomes glutinous and contains threads. After four to five days a whitish pellicle appears on the surface composed of bacteria.
Potatoes.	*Contained in Tubes.*—Striated colonies, yellow or brownish-yellow color. After four to five days gas develops in large bubbles, and the colonies become confluent and at length coalesce. The mass becomes creamy, filamentous, and invades the entire surface of the potato and spreads over the sides.
Temperature.	Best growth at the ordinary temperature—between 30° to 37° C.
Rapidity of growth.	Grows rapidly.
Formation of spores.	Multiply by cleavage.
Aerobiosis.	
Gas production.	Develops numerous bubbles of inodorous gas.
Gelatin reaction.	
Aniline reaction.	Colors with all the aniline stains.
Pathogenesis.	A small quantity of urine containing the organism, mixed with normal urine, becomes acid in reaction after a day and becomes glutinous and thready.

30. Bacillus fluorescens.

Place found.	Water.
Form and arrangement.	Short, fine bacilli with rounded ends.
Motility.	Not motile.
Growth.	
Gelatin.	*Plate Culture.*—Colonies occur on the surface in the figuration of ferns, with a brilliancy resembling mother-of-pearl at the periphery. *Tube Culture.*—Growth very slight along inoculation puncture. More luxuriant on the surface, having the characteristic fluorescence.
Agar-agar.	*Tube Culture.*—Growth superficial, of an emerald-green color.
Potatoes.	Rapid growth, diffusely brown through the body of growth, and the surface assumes a grayish-blue color.
Blood-serum.	
Temperature.	Does not thrive at a high temperature.
Rapidity of growth.	Grows fast.
Formation of spores.	
Aerobiosis.	Does not grow under mica plates.
Gas production.	
Gelatin reaction.	Non-liquefying.
Color production.	Forms greenish-yellow, fluorescent coloring matter.
Pathogenesis.	

31. Bacillus muscoides. *Liborius.* *Zeitschr. f. Hyg., Bd. I, S. 163.*

Place found.	From the œdema fluid of mice inoculated with garden-earth, old cheese, and cow-excrement.
Form and arrangement.	Bacilli about 1 m. thick, with a slight tendency to form threads
Motility.	Slightly motile.
Growth.	
Gelatin.	Easily obtained as soon as anaerobic conditions are produced. *Plate Culture.*—Forms delicate colonies, with slender ramifications, resembling some delicate moss species.
Blood-serum.	
Temperature.	
Rapidity of growth.	
Spore formation.	Of polar origin, round or oval, strongly refractive.
Aerobiosis.	Exquisitely anaerobic.
Gas production.	
Gelatin reaction	Non-liquefying.
Color production.	
Pathogenesis.	

32. Bacillus polypiformis. *See No. 31.*

Place found.	See No. 31.
Form and arrangement.	Thin bacilli, slightly more than 1 m. in thickness, and of various lengths, with no tendency to form threads.
Motility.	Slightly motile.
Growth.	
Gelatin.	Easily obtainable as soon as anaerobic conditions exist. *Plate Culture.*—Growth forms a delicate, yellowish film, with serrated and overlapping periphery. Under low power numerous winding, bending, and intertwining continuations are seen, often polypoid in contour.
Agar-agar.	*On Plates.*—Growth forms whitish colonies of irregular contour, about the size of a pin-head. Under a low power they appear as brownish, finely-granular, mulberry-like masses.
Potatoes.	
Blood-serum.	Forms at the lower portion of the inoculation puncture a diffuse cloudiness.
Temperature.	
Rapidity of growth.	
Formation of spores.	Spores oval and even rod-like, which ofttimes occupy as much as a half and even two-thirds of the parent-cell.
Aerobiosis.	Exquisitely anaerobic.
Gas production.	None in artificial media.
Gelatin reaction.	Non-liquefying.
Color production.	
Pathogenesis.	

33. Bacillus erytrosporus. *Eidam.* *Cohns Beitr. z. Biol. d. Pfl., Bd. III, S. 128. Flügge, Mikroorganismen, S. 288.*

Place found.	In meat-water, egg-albumen, and in drinking-water.
Form and arrangement.	Narrow bacilli, with rounded ends, often forming short threads.
Motility.	Motile.
Growth.	
Gelatin.	*Plate Culture.*—Whitish colonies, which spread over the surface and become plaited and furrowed; around each colony there is a greenish-yellow coloring. Under low power they are found to be circular, with irregular but sharply-defined borders. The opaque, brownish centre is surrounded with a brighter greenish-yellow zone, and the surface shows slightly radiating apophyses. *Tube Culture.*—Luxuriant growth along the puncture; especially so on the surface of gelatin. Under weak illumination the entire gelatin assumes gradually a greenish hue, beginning from above; under stronger illumination the color appears yellowish.
Potatoes.	A thinly-spread layer, at first reddish, later rust-color.
Blood-serum.	
Temperature.	Does not thrive at a high temperature.
Rapidity of growth.	Grows quite rapidly.
Spore formation.	At temperature of room there arises from each bacillus from 2 to 8 oval spores resembling a string of pearls, which, partly divided from the contour of the bacillus, show a distinct, muddy-red color.
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Color production.	Produces a greenish-yellow fluorescence.
Pathogenesis.	

34. Bacillus albus.

Place found.	Water.
Form and arrangement.	Short bacilli with blunted ends. Several individuals are frequently seen lying together.
Motility.	Motile.
Growth.	
Gelatin.	*Plate Culture.*—Round, white colonies, about the size and shape of a pin-head. *Tube Culture.*—Grows slowly along the puncture, of a white color, and on the surface forms a button, as on the plates.
Potatoes.	Growth confined to inoculation stroke of a muddy, yellowish white.
Blood-serum.	
Temperature.	Does not thrive at a high temperature.
Rapidity of growth.	Grows slowly.
Formation of spores.	
Acrobiosis.	Does not grow under mica plates.
Gas production.	
Gelatin reaction.	Non-liquefying.
Color production.	
Pathogenesis.	

35. Proteus Zenkeri. *Hauser. Ueber Fäulnisbakterien. Leipzig. 1885.*

Place found.	In putrefying animal matter.
Form and arrangement.	Bacillus 0.4 m. in breadth and 1.65 m. long. More-rounded forms may occur.
Motility.	Motile.
Growth.	
Gelatin.	*Plate Culture.*—Growth occurs as a thick, whitish-gray coating which may be easily detached. Does not form in zoöglœa, thus differing from Nos. 12 and 13.
	Tube Culture.
Potatoes.	
Blood-serum.	
Temperature.	Thrives best at a temperature ranging from 20° to 24° C.
Rapidity of growth.	Grows fast.
Spore formation.	Spore formation has not been observed, although they withstand desiccation.
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Color production.	
Pathogenesis.	See No. 12.

36. Bacillus of blue milk. *Hüppe.* *Mitth. a. d. Kais. Gesundh.-Amt., Bd. II, S. 355.* *Neelsen.* *Cohn's Beitr. z. Biol., Bd. III, Heft 2.*

Place found.	In blue milk.
Form and arrangement.	Bacilli varying greatly in length, from 1 to 4 m. long, 0.3 to 0.5 m. broad, with slightly-rounded ends.
Motility.	Motile.
Growth.	
Gelatin.	*Plate Culture.*—Finely-seeded, muddy-white, circular colonies, with smooth edges. After some time the gelatin shows a dark discoloration. *Tube Culture.*—Nail-shaped growth with milk-white head; the surrounding gelatin assumes a diffuse, grayish-blue hue, which, later, becomes darker, and lastly black.
Agar-agar.	*Tube Culture.*—Growth of a grayish color, coloring the agar a dark brown.
Potatoes.	Growth of a yellowish color, confined to the inoculation point. The surface assumes a diffuse, grayish blue.
Blood-serum.	Growth characterized by absence of coloring.
Temperature.	
Rapidity of growth.	Grows rapidly.
Formation of spores.	Increases by division and spore formation. The spore conjoined with the bacillus at one end gives a club-shape. Spore formation occurs in the dependent portion of the culture in gelatin, beginning at the third day.
Aerobiosis.	Does not grow under mica plate.
Gas production.	
Gelatin reaction.	Non-liquefying.
Color production.	Produces a grayish-blue coloring matter outside the organism at the expense of the media.
Pathogenesis.	

37. Bacillus lacticus. *Hüppe.* *Mitth. a. d. Kais. Ges.-Amt., Bd. II, S. 337.*

Place found.	In sour milk.
Form and arrangement.	Short, chunky staves, 1 to 1.7 m. in length, 0.3 to 0.4 m. in breadth, arranged usually in pairs, infrequently in chains of fours.
Motility.	Non-motile.
Growth.	
Gelatin.	*On Plates.*—At first the colonies have the appearance of small white dots, which increase to grayish-white plaques, having porcelain-like lustre, the rims of which are transparent. With higher power the superficial colonies have a flat appearance, yellowish in color, with a granular or indented centre, and very delicate, pale, irregular rims. *Tube Culture.*—The growth at first appears evenly distributed along the puncture in the form of delicate dots; and, later, develops on the surface as a dry, grayish-white, shimmering, hemispherical heap.
Potatoes.	
Blood-serum.	
Temperature.	Under 10° C. it is incapable of development in milk. At 10° to 12° C. it commences, and at 15° C. the lactic fermentation is at its height. At 45.5° C. it ceases. In about ten days casein is formed.
Rapidity of growth.	Grows slowly.
Spore formation.	Increases by division and endogenous polar sporulation. Spores are bullet-like in form, glistening, and powerfully refractive.
Aerobiosis.	Indifferent.
Gas production.	
Gelatin reaction.	Non-liquefying.
Color formation.	
Pathogenesis.	*Physiological Attributes.*—Decomposes milk-sugar into lactic acid and carbonic acid, and, by the acidity resulting in the process, casein is formed.

38. Bacillus epidermidis (*Bizzozeros* Lepthothrix epidermidis). *Virchow's Archiv, vol. xcviii, p. 455.* ***Bordoni Uffreduzzi.*** *Fortschr. d. Med., No. 156. 1886.*

Place found.	From scabs and epidermic scales and from the exfoliated particles from between the toes.
Form and arrangement.	Bacilli about 2.8 to 3 m. in length and 0.3 m. in breadth.
Motility.	
Growth.	
Gelatin.	Growth very slight.
Agar-agar.	*Plate Culture.*—Growth occurs on the surface.
Potatoes.	At 15° to 20° C. development begins in the form of drops, which gradually elongate or spread out, and thus coalesce, become more condensed, and form a thin skin over the surface.
Blood-serum.	Growth forms a thin coating.
Temperature.	Best growth produced at 15° to 20° C.
Rapidity of growth.	Grows very slowly.
Spore formation.	Spores are 1.2 to 1.5 m. in length and 0.3 to 0.4 m. in breadth. Sporulation occurs in a distinctly characteristic manner at 25° C. on the third day. At 14° C. there is no formation of spores.
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Color production.	
Pathogenesis.	Inoculations in rabbits and guinea-pigs and on the human skin resulted negatively or failed altogether.

39. Bacterium Zopfii. *Flügge.* *Mikroorganismen, S. 326.*

Place found.	In the intestines of chickens.
Form and arrangement.	Bacilli, 0.75 to 1 m. broad and 2 to 5 m. long; form threads which, on gelatin, are arranged in peculiar and manifold curves.
Motility.	Actively motile.
Growth.	
Gelatin.	*On Plates.*—The colonies resemble those of the spreading mycel fungi. *Tube Culture.*—Along the puncture a compact, yellowish-white line develops, which sends off white filaments which intersect and radiate in every direction.
Potatoes.	
Blood-serum.	No growth.
Temperature.	Thrives best about 20° C. At 30° to 37° C. the motion of the spores ceases. Involution forms arise at a temperature of 37° to 40° C., and if they are exposed for a time to this temperature they die.
Rapidity of growth.	Grows very rapidly.
Spore formation.	Forms spores which were considered cocci by Kurth. They are wanting in some of the peculiarities which are characteristic to spores, *ex. e.g.*, are not strongly refractive, are capable of being stained, and have no resistance against heat and evaporation; but they have the power of developing into bacilli similar to the maternal organisms.
Aerobiosis.	Oxygen is an absolute necessity.
Gas production.	
Gelatin reaction.	Non-liquefying.
Color production.	
Pathogenesis.	They are non-pathogenic to all animals experimented upon.

40. Bacillus resembling bacillus subtilis, I. *Bienstock.* *Zeitschr. f. klin. Med., Bd. VIII, Heft 1.*

Place found.	In fæces.
Form and arrangement.	Bacilli about the size of the hay bacillus, whose ends are rounded. They develop in long threads of from 2 to 5 members; later, the individuals often separate.
Motility.	Non-motile.
Growth. Agar-agar. Potatoes. Blood-serum.	*Tube Culture.*—Growth occurs in a characteristic manner, resembling a mesentery, with veins of a yellowish-white color running in all directions, connected by smaller anastomoses, which later amalgamate.
Temperature.	Grows best at 37° to 39° C.
Rapidity of growth.	
Spore formation.	They possess neither a polar spore formation, nor are they endogenous, as in the bacillus subtilis (No. 14), but the ellipsoid poles of the spores both assume by degrees a cylindrical shape, while the intervening space appears swollen.
Aerobiosis.	
Gas production.	
Gelatin reaction.	
Color production.	
Pathogenesis.	

41. Bacillus resembling the bacillus subtilis, II. *Bienstock.*

Place found.	In fæces.
Form and arrangement.	As in No. 20.
Motility.	
Growth.	
Agar-agar.	*Tube Culture.*—Growth occurs over the surface, is shining and at first smooth, but later becomes uneven. At the periphery there are clusters of bacilli resembling grape-runners in arrangement.
Potatoes.	
Blood-serum.	
Temperature.	Grows best at 37° to 39° C.
Rapidity of growth.	Grows extremely fast. In from ten to twelve hours the surface of the nourishing ground is entirely covered.
Spore formation.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	
Color production.	
Pathogenesis.	

42. Bacillus of albumen decomposition. *Bienstock.*

Place found.	In fæces.
Form and arrangement.	Short and long staves, forming threads, which occupy from $\frac{1}{2}$ to $\frac{2}{3}$ of the microscopic field.
Motility.	Very motile.
Growth.	
Agar-agar.	*Tube Culture.*—Not very characteristic; in the first days it has a shimmer like mother-of-pearl; when allowed to stand some days grows yellow. The surface is homogeneous.
Potatoes.	
Blood serum.	
Temperature.	
Rapidity of growth.	
Spore formation.	Process differs from that of No. 20, and is as follows: Lengthening of the staves into rosary-like chains; separation of the individuals into shorter staves, which increase in length and again form into chains, and so on.
Aerobiosis.	
Gas production.	
Gelatin reaction.	
Color production.	
Pathogenesis.	

43. Bacterium aerogenes. *Miller.* *Deutsche Med. Wochenschr., No. 8. 1886.*

Place found.	From the digestive tract.
Form and arrangement.	Short staves, single and in pairs.
Motility.	Motile.
Growth.	
Gelatin.	*Plate Culture.*—Forms circular, sharply-defined, yellowish colonies, with darker lines radiating from the centre to the periphery, but ending before they arrive at the edges. *Tube Culture.*—Forms along the entire puncture a growth of a brownish-yellow color, and on the surface a flat, grayish-white button.
Agar-agar.	*Tube Culture.*—As a grayish-white, pulp-like coating
Potatoes.	Forms a weak, yellowish-white, pulp-like coating, with irregular edge. (Said to form gas-bubbles.)
Blood-serum.	
Temperature.	
Rapidity of growth.	Grows rather rapidly.
Formation of spores.	
Aerobiosis.	May grow without air, but this hinders their growth. Under cover-glass numerous air-bubbles are formed.
Gas production.	In media containing carbohydrates the production of gas is abundant.
Gelatin reaction.	Non-liquefying.
Color production.	
Pathogenesis.	*Physiological Attributes.*—Possesses great power of resistance against acid, so that it may be exposed for hours to an artificial gastric juice without changing its power of development.

44. Helico bacterium aerogenes. *Miller.* *See No. 43.*

Place found.	From the intestinal tract.
Form and arrangement.	Thin staves, single or in chains, which grow in long, wavy threads, sometimes bent.
Motility.	Motile.
Growth.	
Gelatin.	*Plate Culture.*—Forms transparent, white, often slightly-yellow colonies, which assume a variety of forms; under low power the individual bacilli may be observed composing the threads. *Tube Culture.*—Growth forms evenly along the entire puncture of a light-yellow color; the surface is often covered with a thin, scarcely visible, dry film.
Agar-agar.	*Tube Culture.*—Not especially characteristic.
Potatoes.	Grows slowly. The colonies are of a brownish color, rather dry, whose edges are serrated.
Blood-serum.	
Temperature.	
Rapidity of growth.	
Formation of spores.	
Aerobiosis.	Under anaerobic conditions the growth is very limited.
Gas production	
Gelatin reaction.	Non-liquefying.
Color production.	
Pathogenesis.	See No. 43.

45. Bacillus aerogenes. *Miller.* *See No. 43.*

Place found.	From the digestive tract.
Form and arrangement.	Small staves of different lengths.
Motility.	Motile.
Growth.	
Gelatin.	*Plate Culture.*—Forms exactly round, homogeneous, transparent colonies of a white or light-yellow color; the older ones often possess concentric circles. *Tube Culture.*—Evenly along the puncture, of a yellowish color. On the surface a thin, pearl-gray coating, with scalloped rim.
Potatoes.	Slow growth in form of a dry coating of bluish-yellow, muddy color, and irregular border.
Blood-serum.	
Temperature.	
Rapidity of growth.	
Formation of spores.	
Aerobiosis.	
Gas production.	Same as 44.
Gelatin reaction.	Non-liquefying.
Color production.	
Pathogenesis.	See No. 43.

46. Spirillum rubrum. *v. Esmarch.* *Centralblatt f. Bacteriol. u. Parasitenk, Bd. I, S. 255.*

Place found.	From the putrefaction arising in the dead body of a mouse which had been killed by mice septicæmia. Isolated by the roll method.
Form and arrangement.	Spirillum of from 1 to 3 screw-curves, which in bouillon may develop up to 50. They are about twice the thickness of the cholera spirules.
Motility.	Actively motile.
Growth.	
Gelatin.	*On Gelatin Rolls.*—Growth occurs at room temperature in small colonies, which at first are colored gray or bluish-red, and later wine-red. Their body is granular and their rims smooth. *Tube Culture.*—Growth from the entire puncture. The colonies are round and closely packed. From the first they are of a pretty, wine-red color, with the exception of those lying on the surface at the entrance of the puncture (thus differing from all color-forming organisms).
Agar-agar.	*Tube Culture.*—Forms grayish-white, later pinkish-red, sharply-defined colonies, with a moist, shining surface. They possess no tendency to spread.
Potatoes.	Forms deep-red colonies, varying in size, but usually about that of a hemp-seed.
Blood-serum.	As on agar-agar; in the water expressed in desiccation they form a red sediment. The same thing occurs in bouillon.
Temperature.	Most prolific at 37° C. When let for twenty-four hours at 42° C. the spirules become motionless and lose the power of growth when transplanted.
Rapidity of growth.	Grows very slowly.
Spore formation.	In old cultures on solid media, bright, strongly-refractive spots are observed, which always remain colorless; these must be considered spores, although they have resisted every known method of spore-coloring, inasmuch as cultures consisting only of spirillæ, when dried on threads for six to eight days, are known to redevelop if this spore formation is present. This may take place after even five to eight weeks' exposure to the drying process. They have a poor resistance to heat.
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Color production.	Stains readily with the aniline dyes.
Pathogenesis.	Is non-pathogenic.

II.

PATHOGENIC BACTERIA.

A. Cultivated Outside the Animal Body.

47. Comma bacillus of Asiatic cholera. *Koch.* *Berl. klin. Wochenschrift. 1884. Nos. 31, 32, 33a.*

Place found.	In the intestinal canals of recently moribund cholera patients, and from the fæces of same.
Form and arrangement.	Curved bacilli, about one half or two-thirds, at most, as long as the bacillus tuberculosis. Frequently so connected as to form half-circles or S-shapes. They also form long, delicate screws, resembling the spirilla of relapsing fever in length and appearance.
Motility.	Very motile.
Growth.	
Gelatin.	*Plate Culture.*—Colonies with a more or less irregularly defined, occasionally scalloped contour, of a granulated appearance, resembling pieces of glass. Young, superficial, flat colonies have a delicate, rosy-red tinge. Under a low power liquefaction may be observed in the funnel-shape, and this form is preserved throughout the process. *Tube Culture.*—Liquefaction begins slowly, commencing at the entrance of the puncture around an inclosed air-bubble, and continues along the entire slab in the form of a funnel. After four weeks liquefaction of the entire tube-contents has taken place.
Agar-agar.	*Tube Culture.*—Non-liquefying and not especially characteristic.
Potatoes.	At 30° to 40° C., light, grayish-brown colonies, resembling those of B. mallei, which slowly deliquesce. At room temperature no growth.
Blood-serum.	Rich growth, which liquefies.
Temperature.	Grows best at 30° to 40° C.; under 16° growth seems to cease, though they may endure 10° C.
Rapidity of growth.	Grows rapidly.
Formation of spores.	Forms arthrosporea (Hüppe), but it has no form which possesses a higher measure of resistance.
Aerobiosis.	Does not grow under mica plate.
Gas production.	
Gelatin reaction.	Liquefying.
Aniline reaction.	It is best colored with aqueous sol. of fuchsin, and may be decolorized by Gram's method. *Color Reaction.*—Add from 5 to 10 per cent. of hydrochloric acid to a bouillon culture, growing at 37° C., and a pink or violet color develops, with a brownish tinge in a bright light (Bujwid). Brieger has allowed the same to stand, with the addition of sulphuric acid neutralized with soda and extracted with benzol, thereby producing a Burgundy-red color, which is innocuous—the so-called "cholera-red."
Pathogenesis.	*Experiments on Animals.*—Injections into the stomachs of guinea-pigs of pure cultures in bouillon, with the addition of tr. opii and Na_2Co_3, were successful (Koch). The first attempt at injection into the small intestine, after ligature of the intestine below the bile-duct, were also successful (Nicati and Retsch). The bacilli have been found in the contents of the intestine, the tubular glands, and in the mucous membrane of the intestine. Not as yet found in the blood of man, but positively in the intestinal walls.

48. Finckler-Prior bacillus. *Centr.-Bl. f. allg. Ges.-Pfl., Bd. I, Heft 5, u. 6.*

Place found.	First found in fæces after standing fourteen days.
Form and arrangement.	Somewhat larger than Koch's comma bacillus, but not so curved; growing in spirilli that are not so long as those of the Koch comma bacillus; they are thicker in the middle than at the ends.
Motility.	Motile.
Growth.	
Gelatin.	*Plate Culture.*—Round colonies, having a granular appearance and of a yellowish color, increasing quickly in size and liquefying the media. After twelve to twenty-four hours the entire plate is liquefied and the colonies commingled. *Tube Culture.*—The growth liquefies from the top surface as well as along the puncture, thus forming the so-called stocking-shaped growth; in from one to two weeks the entire contents of the test-tube is liquefied.
Agar-agar.	*Tube Culture.*—Does not liquefy and is without special characteristic.
Potatoes.	Growth at room temperature is not confined closely to inoculation puncture. The colonies assume the form of soft, gravish-yellow plaques, with scalloped edges, and of slimy, glutinous consistency.
Blood-serum.	Growth unusually energetic and liquefying.
Temperature.	
Rapidity of growth.	Growth more rapid than cholera bacillus and the cheese spirillum.
Spore formation.	
Aerobiosis.	Does not grow under mica plate.
Gas production.	
Gelatin reaction.	Liquefying.
Aniline reaction.	Is best colored by aqueous solution of fuchsin. After treatment with HCl a reaction like that of the cholera bacillus is developed, but having a more brownish tinge, and this only after a much greater length of time. (Bujwid.)
Pathogenesis.	Guinea-pigs die after the injection of pure cultures per os, as well as after mixed alkaline cultures have been injected into the stomach or duodenum. The increasing of the bacilli is unusually large in the alimentary canal of the guinea-pig, and are excreted in the urine. (Finckler-Prior.)

49. Cheese spirillum. *D. med. Wochenschr. 1885. No. 3.*

Place found.	In old cheese.
Form and arrangement.	Crooked bacillus, smaller than those of Asiatic cholera, having a tendency to form spirilla threads which acquire the same length as those of Koch's bacillus; but other screw-worms are closer together and of the same diameter along their entire length.
Motility.	Motile.
Growth.	
Gelatin.	*Plate Culture.*—Under low power circular colonies with regular dark contour and greenish-brown centres, irregularly granular. Later, when liquefaction occurs and the colonies sink, the dark color disappears. *Tube Culture.*—The liquefaction tube is larger than that of the cholera bacillus, and occurs as rapidly at the surface as at the end of the inoculation stab.
Agar-agar.	*Tube Culture.*—Growth as a thin, yellowish coat, without special characteristics.
Potatoes.	No growth at room or breeding temperature.
Blood-serum.	Growth of powerful liquefying powers.
Temperature.	
Rapidity of growth.	Growth more rapid than the cholera bacillus and slower than Finckler-Prior bacillus.
Spore formation.	
Acrobiosis.	Does not grow under mica plate.
Gas production.	
Gelatin reaction.	Liquefying.
Aniline reaction.	Is best colored by aqueous solution of fuchsin.
Pathogenesis.	Guinea-pigs are killed by inoculation per os. (Of 15 animals 3 died.)

50. Bacillus cavicida (Brieger's bacillus). *Berl. klin. Woch. 1884. No. 14.*

Place found.	In fæces and in artificial putrefying matter.
Form and arrangement.	Small staff, twice as long as broad.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Very characteristic, occurring in concentric rings, prettily grouped, and of a whitish color. Arranged like the scales of the back of a tortoise.
Potatoes.	Forms a muddy-yellow coating.
Blood-serum.	Thrives well on sterilized human blood.
Temperature.	
Rapidity of growth.	
Formation of spores.	
Aerobiosis.	
Gas production.	Decomposes sugar solutions into hydrocyanic acid, with a trace of acetic acid, thus placing it in the category of putrefaction bacteria.
Gelatin reaction.	
Aniline reaction.	
Pathogenesis.	Very malignant to guinea-pigs, while mice and rabbits often resist its action. A few hours after subcutaneous injection guinea-pigs become unusually quiet, lose their voracious appetites, and succumb within three to twenty-four hours. Bacilli are found in the blood. Per os and per anum they have no effect.

51. Bacillus neapolitanus (Emmerich's bacillus). *D. med. Wochen. 1884. No. 50. Berl. klin. Wochenschr. 1885. No. 15.* **Weisser.** *Zeitschrift f. Hygiene, Bd. I, S. 315.*

Place found.	In stroke-cultures grown from the blood, the juice squeezed from the organs and the discharge from the bowels of those dead from, or attacked with, cholera in Naples. Weisser claims that they are present in normal as well as abnormal human excrement, in air, and putrefying liquids.
Form and arrangement.	Short staves, with rounded ends, single or in pairs, seldom several together. Their length is one and a half times as great as their breadth. In form and size they resemble the typhus bacillus.
Motility.	Non-motile.
Growth.	
Gelatin.	*Plate Culture.*—Occurs as a glassy, milky-white, transparent growth, markedly superficial, and never liquefying. Under a low power those deeper colonies have a whetstone-shape; with strong illumination they appear to have a fine, seeded appearance, and under a weaker illumination they are of a yellowish-brown color. Those on the surface resemble smooth, round mollusk-shells, slightly yellow in the centre, white toward the circumference. *Tube Culture.*—Similar to the typhus bacillus. Possesses the power of increasing the alkalinity of the nourishing media, and thereby causing a cloudiness of the same.
Agar-agar.	*Tube Culture.*—As a whitish, moist coating.
Potatoes.	Forms a yellowish-brown, viscid layer.
Blood-serum.	
Temperature.	Endures exposure to 24° C. for twelve days, and four weeks' desiccation at room-temperature without detriment.
Rapidity of growth.	
Spore formation.	Spore formation not observed. On the other hand, the bacilli have the power of remaining alive for a long time and under very adverse circumstances.
Aerobiosis.	After all oxygen has been consumed they still retain life.
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Is not colored by Gram's method.
Pathogenesis.	Introduced in large numbers into the animal organism (guinea-pigs, dogs, cats, and a monkey), it sets up an inflammation along the small intestine, which presents in all particulars great similarity to human cholera, the duration of which, five to six days, or, more rapid, sixteen to thirty hours, is governed by the number of bacilli introduced. *Sections.*—Spleen normal, great ecchymoses in the cæcum and large intestine, mesentery glands swollen and yellowish. Bacilli are found in all organs and tissues. Weisser could not cause death of animals regularly, and, when it did occur, vomiting and cramps were absent, and the alvine discharges were not liquid, but pap-like.

52. Bacillus diphtheriæ. *Loeffler.* *Mitth. a. d. Kais. Ges.-Amt., vol. ii, p. 421.*

Place found.	In diphtheria membrane.
Form and arrangement.	Straight or crooked staves the same length as the B. tuberculosis, but twice as thick. Stain most intense at the ends (dumb-bell forms).
Motility.	Non-motile.
Growth.	
Gelatin.	*Tube Culture, 5 per cent.*—Between 20° to 22° C., growth takes place as small, round, white dots. The staves assume a dumb-bell form.
Potatoes.	No growth.
Blood-serum.	*On Peptonized Meat-Sugar-Serum Infusion.*—At 37° C. a luxuriant growth occurs, which in two days is nearly 1 mm. thick. There may be isolated colonies, ½ cm. in diameter, or they may form a coating.
Temperature.	A temperature above 20° C. is requisite. Cultures kept for seven weeks at 37° C. still retain their life-power.
Rapidity of growth.	Rapidly on meat-peptone, sugar-serum infusion.
Formation of spores.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	
Aniline reaction.	*Sections are Best Colored by Loeffler's Method.*—A few minutes in 30 c.cm. concentrated alcoholic sol. of methylene blue, to which has been added 100 c.cm. of KHO solution (1 to 10,000), then a few seconds in ¼-per-cent. acetic acid, then absolute alcohol; clear in cedar-oil, mount in balsam.
Pathogenesis.	*Experiments on Animals.*—Mice are refractory; guinea-pigs and birds die after subcutaneous inoculation in a few days. Hæmorrhagic infiltration of the subcutaneous tissue and of the pleuræ are found. When tracheal injection is practiced in chickens, pigeons, and rabbits, a pseudo-membrane is formed.

53. Bacterium coli commune. *Escherich.* *Die Darmbakterin des Säuglings und ihre Beziehungen z. Physiol. der Verd. 1886. Fortschritte der Med. 1885. No. 17.*

Place found.	Constant in the alimentary tube of man, and in those of all the lower animals examined. Especially plentiful in the lower intestine and in the faces of infants.
Form and arrangement.	The typical form is a short, delicate staff 0.4 m. broad, 2 to 3 m. long. The lengths vary, noticeably, however, oval and coccoid forms having been observed, but the staffs predominate, arranged closely together and in pairs.
Motility	Slightly motile.
Growth.	
Gelatin.	*Plate Culture.*—Growth on the surface, arranged in anisometric, flat colonies, of a faint white color, the edges of which are sharply irregular. The surface presents indifferent pictures. *Tube Culture.*—Heavy growth along the inoculated punctnre, in the form of white globules. On the surface this spreads out in a delicate growth resembling that on plates.
Potatoes.	Light, pea-colored, succulent, shining colonies, quite luxuriant, spreading over the entire surface.
Blood-serum.	Forms a white coating.
Temperature.	Thrives better at temperature of the room than at body temperature.
Rapidity of growth.	Grows with moderate rapidity.
Spore formation.	Spore formation not observed. There are suggestive uncolored spots not infrequently observed in the interior of the staffs.
Aerobiosis.	Aerobic. Can develop, however, on nourishing gelatin to which has been added grape-sugar, without addition of acid.
Gas production.	Produces, by anaerobic growth, a gas composed of carbonic acid and water.
Gelatin reaction.	Not liquefying.
Aniline reaction.	
Pathogenesis.	Rabbits, and especially guinea-pigs, succumb to subcutaneous or intra-venous injections of small quantities in from one to three days, with diarrhœa and collapse. The upper intestine presents changes varying from rosy hyperæmia to intense inflammation. Reddened plaques are found, which are markedly infiltrated, and there is sometimes a serous exudate into the peritoneum. In the blood and in the parenchyma of the various organs plentiful quantities of bacilli are found. They are not pathogenic to mice.

54. Bacterium lactis aerogenes. *Escherich.* *See No. 53.*

Place found.	In the small intestine of man, especially of infants and animals fed upon milk; in less numbers in fæces; once found in milk.
Form and arrangement.	Short, thick staves, with rounded corners, 0.5 m. to 0.8 m. broad and 1 to 2 m. long, lying most frequently in pairs, at other times in irregular heaps.
Motility.	Non-motile.
Growth.	
Gelatin.	*Plate Culture.*—Forms on the surface isodiametric, peaked, soft, glazed colonies of a porcelain-white color. The deeper ones form yellowish, round spheres. *Tube Culture.*—Luxuriant growth along the puncture, with a more extensive growth at the entrance of same (nail-form).
Potatoes.	Forms white, luxuriant colonies, interspread with gas-bubbles. They sometimes assume a creamy character.
Blood-serum.	Forms a prominent, moist, shining, white coating.
Temperature.	Thrives at room-temperature, but better at temperature of the body.
Rapidity of growth.	Grows rapidly.
Formation of spores.	See No. 53.
Aerobiosis.	Aerobic. On milk and grape-sugar solutions they are anaerobic.
Gas production.	Grown anaerobic on milk and grape-sugar solutions, it produces a gas composed of carbonic-acid gas and water.
Gelatin reaction.	Non-liquefying.
Aniline reaction.	
Pathogenesis.	See No. 53. *Physiological Attributes.*—Affects albumen only slightly. They are energetic in setting up the lactic-acid fermentation in milk.

55. Bacillus of intestinal diphtheria in rabbits. *Ribbert.* *Deutsche Med. Wochenschrift, 1887, p. 141.*

Place found.	From the organs of rabbits dead of spontaneous intestinal diphtheria.
Form and arrangement.	Staves 3 to 4 m. long, 1 to 1.4 m. thick, with rounded ends, often arranged in pairs or more, forming chains.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Forms light, transparent, gray (later, brownish) colonies, the surface of which is granular and glistening like the mother-of-pearl. *Tube Culture.*—Minimum growth.
Agar-agar.	*Tube Culture.*—Forms a coating which gradually spreads, and which has an indistinct lustre.
Potatoes.	As a white, flat, slowly-spreading coating.
Blood-serum.	
Temperature.	Thrives best at temperature of body.
Rapidity of growth.	Grows quite rapidly.
Formation of spores.	
Aerobiosis.	A minimum growth if no oxygen is allowed to come in contact with the culture.
Gas production.	
Gelatin reaction.	Does not liquefy.
Aniline reaction.	Stains with aniline-fuchsin-water; very difficult to stain otherwise; not at all by Gram's method.
Pathogenesis.	After injection into the vein of a rabbit's ear, death supervenes after three days; on section, large numbers of the bacilli are found in the capillaries of the liver and spleen. Injected subcutaneously, swelling of adjacent lymph-glands follow. Injected into the mouth, they cause an inflammation of the small intestine with superficial necrotic changes, which bear a close resemblance to the diphtheritic process in man, and this before any other symptoms are noticed.

56. Bacillus diphtheriæ columbarum. *Loeffler.* *Mitth. a. d. Kais. Ges.-Amt., vol. ii, p. 482.*

Place found.	In the exudate from a dead pigeon which had succumbed to a disease similar to human diphtheria.
Form and arrangement.	Bacilli somewhat longer and narrower than bacillus of rabbit septicæmia, rounded at the ends, most frequently arranged in heaps.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Colonies beneath the surface form white spheres; on the surface a white coating, which, under low power, gives a yellowish-brown, dusky appearance.
Potatoes.	Forms a coating which varies only slightly in color from that of the potato.
Blood-serum.	Forms a grayish-white, slightly transparent coating.
Temperature.	Grows at room temperature.
Rapidity of growth.	Grows slowly.
Spore formation.	Not known.
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	
Pathogenesis.	Pigeons, sparrows, and rabbits are susceptible; mice peculiarly so, owing to peculiarity of the liver. Chickens, guinea-pigs, rats, and dogs are immuned.

57. Bacillus necrophorus. *Loeffler. Mitth. a. d. Kais. Ges.-Amt., vol. ii, p. 493.*

Place found.	Obtained on the occasion of implanting a broad condyloma in the anterior chamber of the eye of a rabbit.
Form and arrangement.	Bacilli of different lengths but equal breadths, often forming long, thin, slightly wavy threads.
Motility.	
Growth.	
Gelatin.	No growth.
Agar-agar horse blood-serum.	Growth slight.
Neutralized rabbit bouillon.	There arises a white, downy coating, so that the particle of the organ used in inoculation appears to be wrapped in cotton. After some days numerous flakes float in the bouillon, which consist of closely-matted threads of full-grown bacilli.
Temperature.	
Rapidity of growth.	Grows slowly.
Spore formation.	Forms no spores.
Aerobiosis.	
Gas production.	
Gelatin reaction.	No growth on gelatin.
Aniline reaction.	Stains with the commonly employed aniline colors.
Pathogenesis.	Rabbits died after inoculation in the ear or anterior chamber of eye after eight days; at the point of inoculation a necrotic, cheesy process was found. White mice succumbed in six days; at the point of inoculation greenish-yellow, fatty masses were observed. The same were found in the muscles of the back and thigh, in the peritoneum, and everywhere accompanied by the bacilli.

58. Gonococcus. *Neisser.* *Bumm, Der Mikroorganismus der gonorrhoischen Schleimhauterkrankungen. A. Neisser, Centr.-Bl. f. d. med. Wissensch. 1879. No. 28. D. med. Wochenschr. 1882. S. 279.*

Place found.	In the secretions from gonorrhœic inflammation.
Form and arrangement.	Diplococcus, biscuit-shaped, lying in pairs, with flat surfaces lying together, resembling the coffee-bean.
Motility.	Rotatory and oscillatory motion, but no auto-locomotion.
Growth.	
Gelatin.	*Plate Culture.*—Doubtful.
Potatoes.	*Tube Culture.*—Doubtful.
Human blood-serum.	The growth forms a very thin, grayish-yellow coating, which has a moist, smooth surface, the edges of which diffuse gradually into the surrounding media, which is *not* liquefied. Throughout its course it shows a tendency to form jagged protuberances or excrescences, which, with their sharply defined edges, give the appearance of plateau-like mountains, or of an island with precipitous shores.
Temperature.	Grows at 33° to 37° C. Minimum, 25° C.; maximum, 38° C.
Rapidity of growth.	Very slowly.
Formation of spores.	
Aerobiosis.	Increases by fission at right angles.
Gas production.	
Gelatin reaction.	No growth on gelatin.
Aniline reaction.	Fuchsin or methyl violet is best. Is not discolored by Gram's method (Roux).
Pathogenesis.	Inoculation of the urethra of woman with a pure culture on blood-serum produced a severe gonorrhœa (Bumm). Cocci are found in large numbers, nearly always within pus-cells, seldom free.

59. Trachoma coccus. *Sattler, Michael.* *Knapp-Schwaiger's Archiv für Augenheilkunde, Bd. XVI, S. 367.*

Place found.	From contents of conjunctiva follicles of an inmate of the orphan asylum at Aschoffenburg, suffering from the Egyptian ophthalmia in 1885, during an epidemic of the disease, and in the inclosed follicles of trachoma.
Form and arrangement.	Diplococci, biscuit-shaped, characterized by the minuteness of the partition slit.
Motility.	Rotatory and oscillatory movements; no auto-movement.
Growth.	
	Plate Culture.—In whitish films or flakes.
Gelatin.	*Tube Culture.*—A shining, whitish growth, which, at first, has a superficial coloring of gray. Along the inoculation stab small spherical colonies appear, arranged in the shape of a rosary. Later, the coloring becomes yellowish, and in the shrinking assumes the shape of a tulip.
Agar-agar.	*Tube Culture.*—As a homogeneous, gray, or milk-white deposit.
Potatoes.	Slight growth.
Blood-serum.	Along the inoculation a white band is formed. When a portion is removed on a platinum needle it has a mucoid consistency.
Temperature.	Best growth at body-temperature.
Rapidity of growth.	Grows most rapidly on agar-agar and blood-serum,—more slowly on gelatin.
Spore formation.	
Aerobiosis.	Aerobic as far as known.
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Is stained by all basic aniline colors, but not by Gram's method.
Pathogenesis.	Inoculated subcutaneously and in other various ways, and by direct introduction into the anterior chamber of the eye, produced no results in the rabbit. In the human subject, however, inoculation in the conjunctiva was followed by typical trachoma. The cocci were found in the follicles of the excised conjunctiva after artificial as well as spontaneous occurrence, as well as in one case in which cicatrization had taken place. They are situated in and between the cells, most frequently in the centre of the follicles.

60. Bacillus of conjunctivitis. *Koch, Kartulis.* *Koch, Berichte aus Ägypten an der Preussischen Staatmunster d. Innern. Kartulis, Cent. f. Bac., vol. i, p. 289. Knapp-Schweiger's Archiv für Augenheilkunde, Bd. XVII, S. 318.*

Place found.	In the secretions of the conjunctival sac of those affected with conjunctivitis. Found in Alexandria.
Form and arrangement.	Size, form, and arrangement similar to the bacillus of mouse-septicæmia.
Motility.	
Growth.	
Gelatin.	No growth except the inoculation be carried over from agar-agar; even then very slight.
Agar-agar, 0.5 per cent.	*Tube Culture.*—At 28° to 30° C., after thirty to forty hours, fine, small dots of a gray-white color develop along the inoculation stroke. Later, these dots become confluent and form a dark coating, which becomes heaped above the level of the agar, and possesses a shining, fatty appearance. The edges are irregular, undulating, often jagged; old cultures resemble a *fish-bone.*
Potatoes.	A feeble growth.
Blood-serum.	Meat extract furnishes a very good nourishing media. (Kartulis.)
Temperature.	Grows only at 34° to 37° C.
Rapidity of growth.	Grows very slowly.
Spore formation.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	
Aniline reaction.	Is colored slightly by watery solution of aniline, but not at all by Gram's method.
Pathogenesis.	Inoculated on the conjunctiva of man it produces a conjunctivitis. The bacilli were found in the yellowish discharge as well as on the portion of the conjunctiva excised. The experiments were not made with pure cultures: these were always mixed with a club shaped bacillus very difficult to isolate. However, with pure cultures of the latter no results were obtained.

61. Bacillus of Xerosis. *Colomiatti.* *Kischbert-Neisser, Brsl. Aerztl. Zeitschr. E. Fränkel und Franke, Knapp-Schwaiger's Archiv für Augenheilkunde, Bd. XVII, S. 176.*

Place found.	From xerotic masses of the conjunctiva of a child afflicted with xerosis, complicated with keratomalacia and an advanced general atrophy, and in certain forms of conjunctivitis, accompanied by a mucous secretion and hypersecretion of the Meibomian glands.
Form and arrangement.	Bacilli as long as those of mice septicæmia, grouped in large masses. The breadth varies according to the method of staining adopted, which, according to Neisser, is due to a fatty substance in the enveloping membrane.
Motility.	Non motile.
Growth.	
Gelatin.	No growth.
Agar-agar.	*Tube Culture.*—The bacilli were not taken direct from the eye, but were first grown on blood-serum. At 31° to 39° C., a thin mucoid coating arose, giving the surface a greasy appearance.
Potatoes.	No growth.
Blood-serum.	Rosette-shaped colonies develop on both sides of the inoculation stroke for a distance of from 2 to 3 mm., having a pale-gray, greasy appearance; in the condensation water delicate gray scales are visible, which consist of colonies from the surface.
Temperature.	Grows only at body-temperature.
Rapidity of growth.	Grows rapidly in blood-serum.
Formation of spores.	Spherical enlargement at both poles of the bacillus, the body of which colors weakly (Neisser). Propagation also by means of splitting.
Aerobiosis.	
Gas production.	
Gelatin reaction.	No growth.
Aniline reaction.	Preparations which are dried by heat and stained by watery solutions of aniline make the bacilli appear twice as broad as those which are treated with ether and stained with alcoholic solution of aniline.
Pathogenesis.	

62. Bacillus of chicken-cholera (cholera des poules). *Pasteur.*

Place found.	Poultry affected with chicken-cholera.
Form and arrangement.	Short staves, with slightly rounded ends; they color characteristically and seem to often grow in chains.
Motility.	Non-motile.
Growth.	
Gelatin.	*Plate Culture.*—Small, round, white, superficial, granular colonies with uneven borders. *Tube Culture.*—Growth lies on the surface in the form of a delicate, white, scalloped film. No liquefaction.
Agar-agar.	*Tube Culture.*—As a white, shining coating of medium firmness.
Potatoes.	At the temperature of the room there is no growth, but at temperature of the body a scanty, yellowish-gray, transparent tuft arises after some days.
Blood-serum.	As on agar-agar.
Temperature.	At common and breeding temperature.
Rapidity of growth.	Grows very slowly.
Formation of spores.	Not known.
Aerobiosis.	Does not grow under mica plate.
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	The poles color more intensively than the body with aniline colors, giving the impression of a diplococcus or a dumb-bell form of bacillus. In excised preparations they usually accept nuclear staining. Gram's double stain is unsuccessful.
Pathogenesis.	Inoculation of the most minute quantities brings forth in the chicken a characteristic chain of symptoms. They allow the wings to drop; the feathers appear ruffled and dull. They are overcome with drowsiness, and death supervenes within twenty-four to thirty-six hours. At the section hæmorrhagic duodenitis is found, and the bacilli are present here as well as in the visceral organs and blood. Pigeons, sparrows, pheasants, mice, and rabbits are likewise susceptible. Guinea-pigs, sheep, and horses have abscesses at the point of inoculation.

63. Bacillus of rabbit-septicæmia. *Koch.*

Place found.	In drainage water.
Form and arrangement.	
Motility.	
Growth.	
Gelatin.	
Agar-agar.	
Potatoes.	
Blood-serum.	
Temperature.	
Rapidity of growth.	
Spore formation.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	
Aniline reaction.	
Pathogenesis.	

Resembles No. 62. They are very likely identical.

64. Bacillus of swine-plague. *Loeffler, Schütz.* *Arbeiten a. d. Kais. Ges.-Amt., vol. i, Nos. 46 and 376.*

Place found.	In the organs of swine killed by swine-plague.
Form and arrangement.	Oval form, 0.2 m. long, 0.4 to 0.5 m. broad, lying singly or in threads, very similar to the bacillus of rabbit septicæmia.
Motility.	
Growth.	
Gelatin.	*Tube Culture.*—Growth takes place along the stroke in the form of small, gray or white, dim, dot-like colonies, which soon coalesce. At the point of entrance of the puncture a grayish-white tuft arises, whose surface has a shining appearance.
Potatoes.	Doubtful.
In peptone beef infusion.	A dense cloudiness is developed, and, later, a grayish-white, tenacious sediment settles to the bottom of the bulb.
Blood-serum.	Growth in form of a dry, iridescent coating. In the water of condensation a luxuriant growth occurs in the form of grayish, cloudy, mucoid layers.
Temperature.	Grows best at 35° C.
Rapidity of growth.	
Spore formation.	Propagates by splitting.
Aerobiosis.	Facultative aerobic.
Gas production.	Not observed.
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Colors like 62 and 63, so that between the intensely-colored ends the body, for a space of about $\frac{1}{3}$ to $\frac{1}{2}$ their length, remains uncolored. With Weigert's picro-carmine-gentian-violet they assume an intense blue color; they also color well with Gram's method.
Pathogenesis.	They quickly and surely kill mice and rabbits when injected subcutaneously. Introduced into the lungs of a pig, either by directed injection or by respiration, a malignant, infectious pneumonitis is produced.

64*. Bacillus of buffalo-plague. *Oteste, Armann.*

Annali d'Agricoltura. Atti della Commissione per le malatte dagli animali. 1887. Vol. 121.

Place found.	In the blood and in the organs, in the yellowish exudate, in the saliva, bile, fæces, urine, and milk of animals affected with "gray beard."
Form and arrangement.	Bacteria oval, similar to those of rabbit septicæmia, chicken-cholera, and septicæmia of swine. They color more intensely at the ends. They are 0.9 to 1.4 m. longer.
Motility.	They are not actively motile, but possess the oscillatory movements.
Growth.	
Gelatin.	*Plate Culture.*—After two to three days, at ordinary temperature, very small colonies develop, resembling small, round, lustrous drops, which become thinner, granular, and more opaque. The edges are smooth. Their color is pearl-white, which becomes after a time yellowish. *Tube Culture.*—After twenty-four hours the gelatin becomes turbid and of a pearlish-white color along the stab, which becomes more easily seen after two to three days, and is finely granular and transparent.
Agar-agar.	As in gelatin.
Potatoes.	No development.
Blood-serum.	Along the track of the needle a minute, muddy granulation develops, closely arranged and numerous, and of a pearl color. The growth is much slower than in gelatin.
Bouillon.	Ordinary temperature, after twenty-four hours, a cloudiness develops, which after two days is more marked. At the third day the medium becomes clear again, and a muddy-yellowish, flaky sediment is deposited.
Temperature.	Develops at ordinary temperature (16° to 22° C.), better at the temperature of the incubator (37° to 40° C.).
Rapidity of growth.	Moderately slow.
Formation of spores.	The bacteria elongate to double the usual size and separate in the direction of the lesser diameter. The two swollen poles divide and become micrococci, which afterward elongate to bacilli. The stage of sporulation has not as yet been observed.
Aerobiosis.	Does not develop under mica plate, and the growth becomes more vigorous the freer the access of oxygen.
Gelatin reaction.	Does not liquefy gelatin.
Aniline reaction.	It colors well with an aqueous solution of fuchsin, methylin blue, and methylin violet. Beautiful preparations are obtained by Loeffler's method.
Pathogenesis.	*Clinical Picture.*—The animals droop and isolate themselves. They do not eat, grind their teeth, heads droop, and their glance is fixed. Temperature is high. Yellowish mucus runs from nose. Tumefaction of the throat or various other portions of the body. Dyspnœa. Meteorism. In ten to twenty-four days the animal dies in cramps and convulsions. *Section.*—Yellowish, glutinous exudate in the subcutaneous tissue. Considerable injection of the peritoneum and gastro-intestinal mucous membrane. Tumefaction of liver, and hyperæmia of the lungs, which are also slightly œdematous. Abundance of bacteria are found in the blood and tissues. *Experiments.*—Subcutaneous inoculation of male and female buffalo calves, swine, fowls, guinea-pigs, chickens, pigeons, and small birds were followed by characteristic symptoms, followed by death. In the blood and organs and at the point of inoculation numerous specific bacteria were found. Dogs and rats are refractory. Manfred mixed grease with pure cultures and obtained a good vaccine.

65. Bacterium of wild plague. *Kitt, Hüppe.* *Kitt, Sitzungsber. d. Ges. f. Morphol. u. Physiol. München. I. 1885. Hüppe, Berl. klin. Wochenschrift. 1886. Nos. 44, 45, 46.*

Place found.	In the blood and œdema fluid of some undomesticated animals dying of wild plague.
Form and arrangement.	Short staves, 2 to 3 times as long as broad, with markedly-rounded ends. About 4 of them are equal to the diameter of a red blood-corpuscle. According to Hüppe they belong to the cocci which only become slightly elongated in process of development.
Motility.	Non-motile.
Growth.	
Gelatin.	*Plate Culture.*—Along the interior they develop in white (on agar-agar, grayish-white) isolated colonies in the shape of small spheres of the size of a pin-head, which, under a low power, appear granular. *Tube Culture.*—Along the inoculation puncture they occur at first in isolated, small colonies, which, later, coalesce into one mass. On the surface white tufts develop about the size of a lentil, which is smooth, almost circular, and elevated above the solid gelatin.
Agar-agar.	*Tube Culture.*—Same as on gelatin, only more transparent and grayish white.
Potatoes.	As grayish-yellow, slightly-prominent tufts.
Blood-serum.	As a delicate, yellowish, slightly-iridescent layer.
Temperature.	Grows best at breeding-temperature. Under 12° to 13° C. growth seems to cease. Often cultures lose their virulence at room-temperature as well as on that of the incubator.
Rapidity of growth.	Grows rapidly.
Spore formation.	Fructification does not occur by endogenous spore formation. However, a process resembling arthrosporation is observed.
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Cover-glass preparations are easily stained by aqueous solutions of fuchsin, methyl-violet, methyl-blue, and vervain. Sections are best stained in alcoholic solutions of methyl-blue and fuchsin.
Pathogenesis	Pathogenic to all cattle and horses, wild and domestic pigs, rabbits, mice, pigeons, and certain small birds. Rabbits, inoculated cutaneously or subcutaneously or by feeding, die in from fifteen to twenty hours. The bacteria are found almost exclusively in the plasma, and only occasionally within the cellular elements. The epidemic may take the form of a septicæmia with slight œdema and a continuous hæmorrhage, but also may occur as an intestinal mycosis or contagious pleuro-pneumonia. Hüppe considers wild plague, swine-plague, as well as rabbit-septicæmia and chicken-cholera, as different forms of a single infectious disease (septicæmia hæmorrhagica), since cultures from all of these diseases have no distinguishing characteristics. (Hüppe.)

66. Bacillus septicus agrigenus. *Nicolaier.*

Flügge, Mikroorganismen, Leipzig, 1886, p. 257.

Place found.	From field-earth, fertilized with manure.
Form and arrangement.	Bacillus like No. 62 and 63, or somewhat longer.
Motility.	
Growth.	
Gelatin.	*On Plates.*—Under a low power the growth consists of circular, finely granular, sharply defined colonies. In the centre a yellowish-brown color, at the circumference a grayish-yellow color, separated by a darker zone; later, the colors are destroyed, but the granular appearance becomes more pronounced. *Tube Culture.*—As a thin, hardly-characteristic coating.
Potatoes.	
Blood-serum.	
Temperature.	Grows at room-temperature.
Rapidity of growth.	Slowly.
Formation of spores.	
Aerobiosis.	Grows best on surface of gelatin.
Gas production.	Not observed.
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Give the same color reaction as No. 63, only not so frequent or so well defined.
Pathogenesis.	Mice inoculated subcutaneously and rabbits inoculated in the median vein of the ear die in from twelve to twenty-two and twenty-four to thirty-six hours, respectively. In the blood of the heart and of the capillaries of all organs the organisms are found, but only in relatively small numbers. They are frequently found lying close to the blood-disks.

66*. Bacillus of senile gangrene. *E. Tricomi.* *Tricomi, Il microparasita della gangrena. Napoli, 1886. Att. della Soc. Italiana d. chirurgia. Seduta del 20 April, 1887.*

Place found.	In parts attacked by senile gangrene, in blood, gangrenous ichor, in the lymphatic spaces, between the fasciculi of subcutaneous connective tissue. On the proximal side of the line of demarcation they are especially plentiful.
Form and arrangement.	Thin bacilli, not very long, similar to œdema maligna bacillus. Length nearly 3 m., breadth 1 m., ends rounded, lying isolated or in pairs. Some are enlarged at one end (club-form).
Motility.	Non-motile.
Growth.	
Gelatin.	*Plate Culture.*—After twenty-four hours, round, faintly-yellow colonies develop. They are finely granular and have smooth margins, and consist entirely of staves. The gelatin becomes liquefied after thirty-six to forty-eight hours. *Tube Culture.*—After twenty-four hours a slight opacity occurs along the track of the needle, which after forty-eight hours is found to consist of a series of whitish dots. On the fourth day a delicate film occurs on the surface, at the point of entrance of inoculation stroke, which spreads toward the walls of the tube and liquefies in the "funnel-shape," at the apex of which a bubble appears similar to that occurring in the comma bacillus. The gelatin at length becomes clear and the growth deposited at the bottom, which usually consists entirely of spores.
Agar-agar.	After forty-eight hours an irregular white line is seen along the inoculation stroke, along which afterward small colonies develop, which are superficial and, gradually becoming confluent, form a homogeneous film, which is in slight relief.
Bouillon.	After 12 to 24 hours at 37° C., a diffuse turbidity develops, which afterward sinks to the bottom, leaving the media clear.
Potatoe.	After two to three days small white dots appear along the inoculation point which are scarcely perceptible. These spread slowly at ordinary temperature. At 37° C. they become rapidly confluent, forming a dirty-white coating.
Serum.	At 37° C. the development is moderate, identical with that on agar.
Temperature.	Develops well at ordinary temperature; better and more rapidly at 37° C., in Arsonwal's incubator.
Rapidity of growth.	Develops rapidly.
Spore formation.	The bacilli become enlarged at one end or in the middle, from which egg-shaped spores are developed. They are not colored by the ordinary methods, and are strongly refractive. The spores may be colored by methylene violet after passing the stained cover-glass preparation six times through alcohol-flame. Placed in the incubator at 22° C., they develop to ripe bacilli. They show a powerful resistance to drying.
Aerobiosis.	Aerobic.
Gelatin reaction.	Liquefies gelatin.
Aniline reaction.	May be colored by the aniline colors, especially methylene red and violet. Gram's method is also satisfactory.
Pathogenesis.	The bacilli inoculated into the subcutaneous tissue of animals commonly used for experimentation produce a distressing, morbid process, which is similar to that occurring in man, *e.g.*, the subcutaneous tissues are infiltrated with a fetid bloody serum. The muscles are flaccid and in a state of gray infiltration. Portions of the skin are mummified and afterward become tough and black. The gangrene spreads toward the abdomen, whether the inoculation be made in the thigh or back of the animal. Death ensues in guinea-pigs in two to three days, rabbits in four days, and in house-mice after twenty-four hours. White mice are exempt. A committee of the Surgical Congress, Professor Durante, president, before whom these experiments were repeated, indorsed these observations and results of Tricomi.

67. Bacillus œdematis maligni (vibrion septique). ***Hesse.*** *Deutsche med. Wochenschr., 1885, No. 14. Gaffky, Mitth. a. d. Kais. Ges.-Amt., Bd. I, S. 81. Koch, Ebendaselbst, S. 54. Pasteur, Bull. de l'Acad. de Méd., 1887, p. 793. Liborius, Zeitschr. f. Hygiene, Bd. I, S. 158.*

Place found.	In garden-earth.
Form and arrangement.	Staves, 3.0 to 3.5 m. long and 1 to 1.1 m. broad, joined mostly in triplets, often forming threads 14 to 40 m. in length; they are not so broad as bacillus anthracis, and have rounded ends.
Motility.	Very motile.
Growth.	
Gelatin.	*Plate Culture.*—By applying the method of Esmarch and Liborius, the colonies appear as small, shining, spherical bodies, with liquid contents.
Agar-agar.	*On Plates.*—Colonies form a pale-white or smoky cloudiness, the boundaries of which are illy defined. *Tube Culture.*—Diffuse cloudiness surrounding the inoculation stroke. Growth is much facilitated by the addition of 1- to 2-per-cent. solution of grape-sugar.
Potatoes.	At 38° C. it grows within a boiled potato after a piece of tissue removed from some organ of an animal dying of malignant œdema has been implanted. After several days the potato is permeated by a net-work of the bacilli. (Gaffky.)
Guinea-pig bouillon.	(Under hydrogen.) Commences as a diffuse cloudiness of the entire liquid, without the formation of flakes. Then, after two or three days a whitish precipitate is thrown down, and the liquid becomes clear. Differentiation from anthrax bacillus. (Kitasato.)
Blood-serum.	Diffuse cloudiness along the inoculation stroke.
Temperature.	Thrives best at temperature of the body.
Rapidity of growth.	Grows very rapidly.
Spore formation.	Spore formation occurs in cultures at the end of first day. The bacilli swell at either end, or in the middle, thus assuming a spindle form; at this stage they are capable of staining. Later, a spot appears that cannot be colored, and at last a fragile elliptical or cylindrical spore develops, bluish in color and refractive. Maximum spore formation at 37° C., minimum at room-temperature.
Aerobiosis.	Exquisitely anaerobic. Grows only when oxygen is excluded.
Gas production.	According to Liborius the pure cultures do not form gas, and when this does occur there is contamination. Upon opening a culture in guinea-pig bouillon a very penetrating stink is given off.
Gelatin reaction.	Liquefying.
Aniline reaction.	All the aniline series answer as good coloring agents, and when stained they often present a beaded appearance. Gram's method is useless.
Pathogenesis	Spore-containing garden-earth, when injected into guinea-pigs, causes their death within twelve to twenty-four hours; 0.1 to 0.5 c.cm. of bouillon culture, injected subcutaneously in guinea-pigs or mice, causes death in eight to fifteen hours. *Post-mortem Section.*—Extensive subcutaneous œdema commencing at the point of inoculation. The fluid is reddish, clear, and contains bacilli and isolated bubbles of gas; the skin is tense. Bacilli are found in the fluids of the various organs, and in and on their serous coverings, especially in the ascitic fluid; in the blood of the heart only some time after death. Guinea-pigs inoculated with the contents of serous cavities die rapidly.

68. Bacillus of pseudo-œdema. *Liborius.*

Zeitschr. f. Hygiene, Bd. I, S. 163.

Place found.	In the œdema-fluid and tissues of mice inoculated with garden-earth.
Form and arrangement.	Bacilli thicker than the œdema bacillus, with a distinct, bright fimbria surrounding them.
Motility.	
Growth.	Easily obtained as soon as anaerobic conditions are present.
Gelatin.	*Plate Culture.*—Colonies form spherical bodies, at first the size of a hemp-seed, and later that of a pea, with liquid contents; the lower part containing a whitish sediment, surmounted by a clear fluid. In the centre a small gas-bubble, which gradually increases in size.
Agar-agar (containing sugar).	*Plate Culture* (containing sugar).—Forms spherical, oval, or whetstone-shaped colonies, with irregular contour. *Tube Culture.*—A pronounced cloudiness forms, gas-bubbles develop all through the media, and a clear fluid is expressed.
Temperature.	Grows at room-temperature.
Rapidity of growth.	Grows rapidly.
Formation of spores.	Usually each bacillus forms two spores, without any perceptible change in its appearance.
Aerobiosis.	Exquisitely anaerobic.
Gas production.	Produces gas the odor of which resembles that of old cheese, and probably is due to butyric acid.
Gelatin reaction.	Non-liquefying.
Aniline reaction.	
Pathogenesis.	A rabbit, after inoculation with 0.5 c.cm. of the liquid expressed from an agar culture, died after eight hours. No bacilli in blood or spleen. A mouse, after inoculation with 0.5 c.cm., died in twenty hours. In the spleen no bacilli; in blood of heart isolated masses. Rabbits, after intra-venous injections of 1.5 c.cm., die in forty-five minutes. Section showed fibrinous peritonitis, and the spleen and heart blood contained numerous bacilli. The bacilli seem to be injurious from their ptomaines.

69. Bacillus murisepticus (mouse-septicæmia). *Koch.* *Ueber die Ätiologie der Wundinfektionskrankheiten. Gaffky, Loeffler, Mitth. a. d. Kais. Ges.-Amt., Bd. I, S. 80 u. 135.*

Place found.	In drainage-water and putrid liquids.
Form and arrangement.	Very small staves, 0.8 to 1.0 m. long, 0.1 to 0.2 m. thick, often in pairs. At first sight they greatly resemble acicular crystals.
Motility.	Non-motile.
Growth.	*On Plates.*—They do not form a surface growth, and the colonies only form a diffuse cloudiness.
Gelatin.	*Tube Culture.*—The growth is slow, and is not limited either to the surface or along the inoculation puncture. Its color is white, resembling white clouds, which gradually change to a diffuse opacity. In gelatin of a strong alkaline reaction slight liquefaction occurs.
Agar-agar.	*Tube Culture.*—Definite, yellowish-white colonies.
Potatoes.	No growth.
Blood-serum.	
Temperature.	At room-temperature.
Rapidity of growth.	Grows slowly.
Spore formation.	Forms spores.
Aerobiosis.	Grows under mica plate. Facultative anaerobic.
Gas production.	Not observed.
Gelatin reaction.	Non-liquefying, except in old cultures, and then only superficially.
Aniline reaction.	Bacilli in sections are colored well by the common nuclear stains. Gram's method is also applicable.
Pathogenesis.	Domestic mice inoculated subcutaneously die in from forty to sixty hours. The course of symptoms is ushered in by an increased secretion from the conjunctiva and agglutination of the eyelids, followed by drowsiness, the animal sitting with back arched and coat ruffled. Thus death follows, and the animal is found sitting in the same crouching posture after life has fled. Numerous bacilli are found in the subcutaneous cellular tissue near the point of inoculation, through the entire circulatory system, even to the capillaries of the various organs. They are found inside the leucocytes, where they destroy the cells by multiplication, so that the body of a leucocyte may only be represented by a mass of bacilli. Field-mice are immune.

69*. Bacillus of hæmorrhagic infection. *Tizzoni, Giovannini.*

Ref. Med., August 29, 1888.

Place found.	In the pustules and blood of a female child dying of impetigo contagion, with hæmorrhage.
Form and arrangement.	Length, 1 to 1.3 m.; breadth, 0.2 to 0.4 m. Similar to B. of mouse-septicæmia, but a little broader.
Motility.	Non-motile.
Growth.	
Gelatin.	*Plate Culture.*—In general, similar to strep. pyogenes. After two days, small refractive dots with very irregular borders; after four to five days, round, slightly-raised, grayish-yellow colonies, which under low-power magnification disclose a granular surface, while on the rims are seen fine interlacing of filaments, resembling platted, wavy hair. *Tube Culture.*—At 22° C., after two days, a superficial, whitish-gray point arises at the entrance of the inoculation stab, which increases in size, with pure-white irregularly prominent rim, while the centre is slightly depressed. In the inoculation canal, a streak with jagged edges. They spread far into the media.
Agar-agar.	*Plate Culture.*—Like that on gelatin, only quicker development, and of a paler color; often shows a central thickening, which is darker colored. At the periphery a fine, net-like arrangement of bacilli chains. *Tube Culture.*—After twelve to twenty-four hours, grayish-white, slightly prominent points, which later become confluent.
Blood-serum.	Resembles growth on agar-agar. No liquefaction.
Potatoes.	10° to 12° C. no growth. At 35° C. growth is shown as a dark-yellow coloration, confined to the point of inoculation.
Temperature.	Develops at room temperature and incubator temperature.
Rapidity of growth.	Grows slowly.
Spore formation.	No spores have as yet been observed. The bacilli divide in the direction of their breadth, forming smaller elements.
Aerobiosis.	Facultative anaerobic.
Gas production.	Old cultures develop a specially penetrating odor.
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Colored by Gram's method and by the ordinary aniline colors.
Pathogenesis.	Rabbits and guinea-pigs inoculated with ½ to 1 c.cm. of culture die in one to two days. White mice are refractory. The temperature rapidly arose and as rapidly fell in the animals experimented upon. The hair became rough, anorexia developed. Contraction of the posterior extremities, and a marked diminution of the urinary secretion. *Section.*—Punctiform, hæmorrhagic œdema at the place of inoculation, accompanied by ulceration. Acute nephritis. Inflammation of small intestines, which are distended either with a serous fluid or blood. Acute fatty degeneration of the liver. Punctiform hæmorrhages in the great omentum. In very acute cases, hæmorrhages of the lungs occur. Spleen normal, only a little browner than usual.

70. Bacillus of pneumo-enteritis of the pig. *Loeffler, Schütz.*

Arbeiten a. d. Kais. Ges.-Amt., Bd. I, Heft 1 u. 2.

Place found.	From the spleen of swine affected with pneumo-enteritis.
Form and arrangement.	Very like the bacillus of septicæmia of mice, but somewhat slenderer and shorter.
Motility.	
Growth.	
Gelatin.	*Tube Culture.*—Remarkably like No. 69. The cloudiness is rather more confined to the gelatin immediately surrounding the inoculation stab, is denser and more distinct. (Glass-brush appearance.—Schottelius.)
Potatoes.	
Blood-serum.	
Temperature.	Grows at room temperature.
Rapidity of growth.	Grows slowly.
Formation of spores.	Very probably form spores.
Aerobiosis.	Grows under mica plate.
Gas production.	
Gelatin reaction.	Not liquefying.
Aniline reaction.	Loeffler's potassium-methyl-blue solution and Gram's method are both good.
Pathogenesis.	Inoculated with a pure culture, mice die in two to three days, pigeons in three to four days, rabbits in six days. Guinea-pigs seem to possess an immunity. Pigs fall sick and die after having been inoculated with pure culture. Great numbers of bacilli are found in lung, spleen, liver, kidneys, and lymph-channels, but only a few in the blood.

71. Bacillus of pseudo-septicæmia of mice (coprogenus parvus). *Bienstock.* *Zeitschr. f. klin. Med., Bd. VIII, Heft 1.*

Place found.	In fæces.
Form and arrangement.	Extremely small bacilli, resembling No. 70, but somewhat blunter and thicker; easily mistaken for micrococci.
Motility.	Non-motile.
Growth. Gelatin. Potatoes. Blood-serum.	*Tube Culture.*—After weeks the growth has extended only 1 mm. from the inoculation stroke, and is scarcely visible.
Temperature.	
Rapidity of growth.	Grows very slowly.
Formation of spores.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Not known.
Aniline reaction.	
Pathogenesis.	When inoculated at the base of the ear of mice, an extensive œdema is occasioned at the point of introduction and its surroundings. After twenty-four hours death results. The œdema liquid contains numbers of bacilli, the blood of the heart only a few. Cultures believed to be pure after many generations kill rabbits on the eighth day, preceded by an erysipelatous swelling.

72. Bacillus coprogenes fœtidus. *Schottelius.* *Schottelius, Der Rotlauf der Schweine. Wiesbaden, 1885.*

Place found.	In the alimentary contents of swine.
Form and arrangement.	Similar to bacillus subtilis, but shorter and with rounded ends.
Motility.	Non-motile.
Growth.	
Gelatin.	*Plate Culture.*—Pale, yellowish, circumscribed colonies, forming on the surface a fine, transparent film. A penetrating stench is developed.
Potatoes.	In the form of a dry, light-gray coating.
Blood-serum.	
Temperature.	Grows at room temperature.
Rapidity of growth.	Grows rather rapidly.
Formation of spores.	Forms spores closely arranged together in strings.
Aerobiosis.	Spore formation occurs only by admission of air.
Gas production.	Breaks up the albuminoid molecule and produces a very penetrating gas with a very disagreeable odor.
Gelatin reaction.	Non-liquefying.
Aniline reaction.	
Pathogenesis.	Mice and rabbits withstand subcutaneous injections of small quantities, but in large numbers they are toxic to rabbits, whereas swine are immune.

73. Bacillus anthracis. *Pollender, Davaine, Koch.* *Cohn's Beitr. z. Biol. d. Pflanzen., Bd. II, Heft. 3. Mitth. a. d. Kais. Ges.-Amt., Bd. I.*

Place found.	In the blood of animals afflicted with or dying of splenic fever.
Form and arrangement.	Staves from 3 to 20 m. long and 1.0 to 1.25 m. broad, with sharply-cut ends. They divide when they reach a size double the length of an individual bacillus. In bouillon at 36° C. they form long threads.
Motility.	Non motile.
Growth.	
Gelatin.	*Plate Culture.*—Round, white colonies. Under a low power tangled or interwoven threads are seen at the centre of a colony, and at the periphery these in bundles spread out and resemble locks of hair. They develop in about two days. *Tube Culture.*—A whitish cloud or line develops along the inoculation stroke, from which fine filaments spread out in the gelatin, which frequently intertwine and form a delicate network. Liquefaction begins at the surface.
Agar-agar.	*Tube Culture.*—Characteristic, dry, easily-detached coating.
Potatoes.	A luxuriant growth, in the form of dry, white colonies, confined to the inoculation stroke. Spore formation is quite abundant, and occurs most plenteous at breeding temperature.
Blood-serum.	Liquefaction occurs.
Temperature.	Do not develop under 12° to 14° C., or over 45° C. Their power of development is not lessened by being frozen. (von Frisch.)
Rapidity of growth.	Grows rapidly.
Spore formation.	Spores are egg-shaped, which spring from the long axes of the maternal cells. The maximum temperature at which they may be developed is 43° C., the minimum 12° to 18° C.
Aerobiosis.	Does not grow under mica plate.
Gas production.	Not observed.
Gelatin reaction.	Liquefying.
Aniline reaction.	The usual nuclear stains are applicable in section staining. Grain's method is also good.
Pathogenesis.	Mice, guinea-pigs, and most animals inoculated die in twenty-four hours; the former are usually found lying on their backs. *Post-mortem Section.*—Gelatinous deposits in the subcutaneous connective tissue around the point of inoculation; great enlargement of the spleen. The bacilli are found in the blood and in all organs.

74. Bacillus of symptomatic anthrax. *Feser, Bollinger.* *Arloing, Cornevin, Thomas, Le charbon symptomatique du boeuf. Paris, 1887. Kitt, Centralbl. f. Bakteriol. u. Paras, Bd. I, S. 684.*

Place found.	In the myoma; in the inflammatory, bloody serum of the solid tissues; in the bloody transudations of the serous cavities, and in the gall of animals sick or dying of sympathetic anthrax.
Form and arrangement.	Fine staves 0.01 to 0.015 mm. long and up to 0.003 mm. broad.
Motility.	Rotatory movements.
Growth.	To be had under anaerobic conditions.
Gelatin.	*Plate Culture.*—Columns form irregular spheres with verrucous surfaces. Later, liquefaction occurs in their neighborhood, into which the thread radiates. In higher cultures, at 20° to 25° C., after two to three days, a slightly characteristic growth occurs along the inoculation stroke, accompanied by the formation of gas. *Tube Culture.*—At culture temperature, after twenty-four to twenty-eight hours, growth occurs, accompanied by the formation of gas, which possesses a characteristic acid, penetrating odor. (Kitasato.)
On acid guinea-pig bouillon.	Under hydrogen, at 35° to 38° C., after twenty-four to twenty-eight hours, cloudiness of the entire liquid, and formation of separated flaky bodies, which float around in the bouillon. Gas is developed on the borders of the culture, and, lastly, a whitish precipitate is thrown down.
Temperature.	Like the anthrax bacillus, the virulence of these are modified by degrees of temperature. Exposed for twenty hours to a temperature of 100° C. their virulence is lost, while at so low a temperature as 13° C. no deleterious effects are noticeable. Chemicals also lessen their virulence. Under a temperature of 70° C. for two hours and ten minutes their virulence is much increased, and after two hours and twenty minutes it is lost.
Rapidity of growth.	Grows slowly.
Formation of spores.	Oval spores are formed in solid media and the animal body a short time after death. They are stained by Ziehl's method, not by Gram's.
Aerobiosis.	Exquisitely anaerobic.
Gas production.	Large gas-bubbles are formed in the course of growth on gelatin.
Gelatin reaction.	Liquefying.
Aniline reaction.	Aqueous solutions of aniline colors are quite suitable. An especially pretty effect is produced by double staining with eosin and gentian.
Pathogenesis.	Guinea-pigs may be easily inoculated with sympathetic anthrax from cattle, sheep, or goats. Horses, mules, and white rats only suffer from a local tumefaction, while pigs, dogs, cats, rabbits, ducks, chickens, and pigeons almost always remain immune. Frogs are killed only when placed in water at a temperature of 22° C. Intra-venous and intra-bronchial injection confers an immunity upon the animals so treated.

75. Bacillus pneumoniæ. *Friedlaender.*

Fortschr. d. Med. 1883. S. 715.

Place found.	In the pneumonic lung (croupous).
Form and arrangement.	Oval cocci or quite thick bacilli. The cocci often lie in pairs, with their tapering ends in apposition. Those in the lung are most often inclosed in a capsule.
Motility.	Non-motile.
Growth.	
Gelatin.	*Plate Culture.*—Under the surface, sharply-defined, dark-yellowish, finely-granular colonies, which on the surface assume well-defined, whitish elevations. *Test-tube Cultures.*—After twenty-four hours at room temperature a slight grayish-white growth is visible, which assumes the shape of a round-headed nail. The head is formed first, and takes on a porcelain-like glaze, and assumes the form of a split pea. After a. time the gelatin is colored a light brown, and gas-bubbles are formed.
Agar-agar.	*Tube Culture.*—Growth the same as on gelatin, with a white, porcelain-like color.
Potatoes.	Forms a yellowish, moist, viscid mass, in which gas is produced after a day, especially if the potato is kept at the brood temperature.
Blood-serum.	The growth is gray and slimy.
Temperature.	Best growth at 16° to 30° C.
Rapidity of growth.	Grows rapidly.
Spore formation.	Spore formation has not as yet been observed with certainty.
Aerobiosis.	Grows under mica plate. Facultative aerobic.
Gas production.	Gas formation occurs in 4-per-cent. gelatin, and on potatoes at a temperature of 43° C.
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Sections are colored well by the common nuclear stains. Discolored by Gram's method.
Pathogenesis.	Mice, guinea-pigs, and dogs are affected with pneumonia after the bacilli have been injected into the pleura or have been inhaled; but this result is not constant. Rabbits are refractory.

76. Bacillus pseudo-pneumonicus. *Passet.* *Untersuchungen über die Ätiologie der eiterigen Phlegmone des Menschen.*

Place found.	In pus.
Form and arrangement.	Cocci, occasionally short staves, and *vice versa*, as in the true bacillus of pneumonia.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Small, grayish-white dots, consisting of cocci. *Tube Culture.*—After twenty-four hours, gravish-white, shining, "nail-head," hemispherical in shape, on the surface. Along the inoculation puncture itself, no growth. In three to four weeks a brownish discoloration of the gelatin occurs, which is accompanied by a putrid odor.
Potatoes.	At 37° C., a thick, white, succulent coating develops. Gas is absent.
Blood-serum.	
Temperature.	Grows at room and incubation temperature.
Rapidity of growth.	Grows rapidly.
Formation of spores.	
Aerobiosis.	Aerobic.
Gas production.	Slight. (Compare No. 75.)
Gelatin reaction.	Non-liquefying.
Aniline reaction.	
Pathogenesis.	Injected into serous cavities, the cocci cause an inflammation, accompanied by the formation of pus. A milder effect is observed after subcutaneous injection. No results from inhalatiou.

77. Diplococcus pneumoniæ (microbe of sputum septicæmia). *A. Fränkel, Weichselbaum.* *Mikrobe der Sputumseptikämie. Fränkel, Zeitschr. f. kl. Med., Bd. X, S. 401; Bd. XI, Heft 5 u. 6. D. Med. Wochenschrift, 1884, No. 25.*

Place found.	In the sputum of those suffering from pulmonary disease, and especially rusty sputum of pneumonia; in severe cases of empyema in the exudate of cerebro-spinal meningitis.
Form and arrangement.	Oval diplococci, lance-shaped. By higher power they appear as staves, and their pointed ends are seen to be separated by a small space. They are often joined together in chains of five or six members. In preparations from the body (but never from artificial cultures) they are inclosed in capsules.
Motility.	Non-motile.
Growth.	
Gelatin.	*Plate Culture.*—Difficult to cultivate. On 15-per-cent. gelatin at 24° C., small, round, sharply-defined, slightly-granular colonies, of a whitish color. Their growth is slow. *Tube Culture.*—Along the inoculation stroke a series of small globules develop which are distinctly separated from one another, resembling in this respect No. 97. All cultures are distinguished by their delicacy and the rapidity with which the growths lose the power of reproduction.
Agar-agar.	*Plate Culture.*—At 35° C. delicate, shining, semi-transparent, and exceedingly fine dotlets appear, which are scarcely visible to to the naked eye.
Potatoes.	
Blood-serum.	As a transparent veil resembling dew.
Temperature.	Does not develop under 24° C. Optimum, 35° C. Above 42° C. growth ceases. Its virulence is modified by exposure to different temperatures.
Rapidity of growth.	Grows slowly.
Spore formation.	
Aerobiosis.	Thrives well after addition of carbonic acid.
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Is readily stained by the aniline colors, with the exception of the capsule, which remains uncolored. Gram's method may also be applied.
Pathogenesis.	Pathological to mice, guinea-pigs, and rabbits, which die in from twenty-four to twenty-eight hours. Encapsulated microorganisms are found in great numbers in the blood as well as in all the organs. Very little reaction at point of inoculation; spleen greatly enlarged; kept at temperature below or above that of the body, the virulence of the bacillus becomes diminished. It is identical with the organisms known under the names diplococcus lancolatus (Fra Bardon), streptococcus lancolatus Pasteuri (Gamaneia).

78. Diplococcus intercellularis meningitidis. *Weichselbaum.*

Fortschritte der Medicin. 1887. No. 18.

Place found.	In the fresh exudate of meningitis cerebro-spinalis in six cases.
Form and arrangement.	Cocci, single, more frequently lying in pairs or fours, forming tiny heaps. Often specially large ones are observed; probably these are in the act of division. It is characteristic to find them within the cell.
Motility.	
Growth.	
Gelatin.	No growth.
Agar-agar, with 2-per-cent. gelatin.	*Plate Cultures.*—The deeper-lying colonies are very small; the superficial ones are larger and gray. Under low power the former are found to be round or somewhat irregular, finely granulated, with indented edges, and have a yellowish-brown color; the latter possess a yellowish nucleus with an inner bluish-yellow colored areola, and an outer one that is nearly colorless and more transparent. *Tube Culture.*—Growth ceases at a slight distance from the surface, but there it is luxuriant, forming a smooth, gray, viscid mass, on the confines of which are single, confluent colonies.
Potatoes.	No growth.
Blood-serum.	(On fluid from human hydro-thorax.) Very slight growth, hardly visible, very shallow, colorless, and seeded.
Temperature.	At temperature of room, no growth. This only takes place at incubation temperature.
Rapidity of growth.	Reaches maximum growth after forty-eight hours. The longest time it is capable of successive cultivation is six days. It is best to carry on re-inoculation after two days.
Spore formation.	
Aerobiosis.	Shows a marked tendency to grow upon the surface.
Gas production.	
Gelatin reaction.	No growth.
Aniline reaction.	Sections are best colored by Loeffler's alcoholic methyl-blue solution. They become discolored by Gram's method.
Pathogenesis.	Pathogenic to mice, guinea-pigs, rabbits, and dogs. The former are especially susceptible, death occurring in from thirty-six to forty-eight hours. Multiplication occurs within the organism.

79. Bacillus salivarius septicus. *Biondi.*

Zeitschrift f. Hygiene, Bd. II, S. 196.

Place found.	In saliva of healthy and diseased individuals.
Form and arrangement.	Short, elliptical staves with ends somewhat pointed, and relatively thicker body. In blood and exudates they occur as diplobacilli; in animal tissue they sometimes occur in pairs, inclosed within capsules; at others, they are arranged in chains or small heaps.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Best growth by addition of 0.05 per cent. HCl, or 0.02 per cent. HNO_3 or 0.04 per cent. H_3PO_4. Beneath the surface, small, circular colonies appear, which possess an opalescent centre and a transparent periphery. In structure they consist of a zig-zag net-work. Their contour is sharply marked. *Tube Culture.*—Appears as a bright, thin, very delicate, and evenly-drawn line, with very fine dots at its periphery.
Agar-agar.	*Tube Culture.*—Success attends those cultures made directly from the blood; seldom those carried on from previous artificial cultures. The growth appears on the surface, and forms a fine, transparent covering, having the appearance of dew-drops.
Bouillon.	Without cloudiness.
Temperature.	Optimum temperature, 35° to 37° C. A temperature of 20° to 22° C. is the maximum at which it preserves its virulence, and this after twenty days is spontaneously weakened. At 10° C. no development. At 8° to 9° C. they lose their power of developing in fresh culture and also their virulence.
Rapidity of growth.	Growth extremely slow.
Spore formation.	Appears to have no lasting spores. Propagation occurs by segmentation. It is very susceptible to the drying process.
Aerobiosis.	Grows under glass cover and also in an atmosphere of carbonic-acid gas.
Gas production.	Not observed.
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Is colored by all aniline colors; also by Gram's method.
Pathogenesis.	Pathogenic to mice and rabbits, which die in the course of twenty-four to seventy-two hours and fifteen to thirty days, respectively, and present the clinical picture and anatomical signs of an acute or subacute septicæmia. The most intense effect is to be observed at point of inoculation. Bacilli usually lie in pairs, and always between the blood-corpuscles. Attenuated cultures serve as a true vaccine, which protects those animals inoculated with virus procured from a severe infection. Not pathogenic to guinea-pigs, thus differing from No. 77.

80. Bacillus crassus sputigenus. *Kreibohm.* *Inaugural Dissertation, Gottingen, 1889. Flügge, Mikroorganismen. Leipzig, 1886. S. 260.*

Place found.	From sputum and coating of tongue.
Form and arrangement.	Short, thick staves, occasionally oblong, with rounded ends, often curved and twisted like a sausage.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Forms grayish-white, round, viscid drops, which project above the level of the gelatin. Under high power the youngest colonies are superficial, and possess dark points, or short, dark flourishes, of a brownish color. The more superficial ones are brighter and irregularly circumscribed, and have granular edges. *Tube Culture.*—Develops very rapidly, and presents a typical nail-form growth.
Potatoes.	As a thick, grayish-white, moist, shining coating, resembling a culture of pneumo-bacillus, only somewhat less elevated and tougher.
Temperature.	Grows at room temperature.
Rapidity of growth.	Grows rapidly.
Spore formation.	At 35° C. spore formation begins.
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Easily stained. Gram's method is also suitable.
Pathogenesis	Mice die after inoculation with small quantities in forty-eight hours. In the blood and organs (liver) numerous bacilli are found. After intra-venous injection rabbits are killed in twenty-four hours with septicæmic symptoms. Large masses of bacilli in pure culture produce, in rabbits and dogs, diarrhœa and bloody stools, and death in three to ten hours. Section shows the characteristic signs of acute gastro enteritis.

81. Bacillus oxytocus perniciosus. *Wyssokowitsch.*

Flügge, Mikroorganismen. Leipzig, 1886. S. 268.

Place found.	From milk allowed to stand a long time.
Form and arrangement.	Short staves with rounded ends, somewhat shorter and thicker than the common lactic-acid bacteriæ.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Beneath the surface, small, yellow colonies; under a low power the colonies appear finely granular, sharply defined and circular, of a yellowish-brown color. On the surface the colonies grow until their diameter is 1½ mm.; these are grayish-white, round, and elevated. *Tube Culture.*—At first "nail form;" later, the growth spreads over the entire surface.
In milk.	Causes lactic-acid fermentation, but without odor.
Temperature.	
Rapidity of growth.	
Spore formation.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	
Pathogenesis.	Small quantities inoculated in mice or rabbits have no effect at all. Cultures from one or two test-tubes injected into the ear-vein of rabbits caused death without exception in 6 cases within three to twenty two hours. About three-quarters of an hour after the injection severe diarrhœa ensued. At section, intense or hæmorrhagic inflammation of the intestinal mucous membrane was found.

82. Diplococcus of pneumonia in the horse. *Schütz.*

Virchow's Archiv, Bd. 107, S. 374.

Place found.	In the lungs of horses afflicted with genuine pneumonia.
Form and arrangement.	Oval cocci which divide in the direction of least diameter and lie most commonly in pairs, and possess a bright, homogeneous matrix.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—At room temperature, small, round, white colonies are formed. *Tube Culture.*—Along the inoculation stroke as small, white, disconnected dots, which, although they grow larger, never amalgamate nor project above the surface.
Agar-agar.	*Plate Culture.*—Colonies resembling small, transparent drops.
Potatoes.	
Blood-serum.	No growth.
Temperature.	Grows at room temperature.
Rapidity of growth.	Grows slowly.
Formation of spores.	Not known.
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Are decolorized by Gram's method.
Pathogenesis.	Pathogenous to mice, rabbits, and guinea-pigs. Direct injection into horses' lungs produces death within eight to nine days.

83. Staphylococcus pyogenes aureus. *Rosenbach.* *Mikroorganismen bei den Wundinfektionskrankheiten des Menschen. Passet, Ætiologie der eiterigen Phlegmone des Menschen.*

Place found.	Very frequently in pus, in draff, and in earth. (Ullmann.)
Form and arrangement.	Cocci of irregular size, arranged in heaps; often as diplococci. The medium size is 0.87 m.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—After two days punctiform colonies of a yellowish color, sharply defined, which set in a slight depression in the solid gelatin. *Tube Culture.*—At first the growth appears as a dim, white line along the puncture; in about three days it assumes a yellowish color, and, liquefying the geletin, sinks to the bottom as an orange-yellow deposit.
Agar-agar.	*Tube Culture.*—Non-transparent, yellowish, knotty masses along the stroke. After twenty-four hours a slightly opaque, later a yellowish to orange-colored growth develops, 3 or 4 mm. in breadth, having a wavy edge.
Potatoes.	Thin, whitish coating, which grows more and more succulent, of an orange-color, and having a strong, sour odor.
Blood-serum.	As on agar-agar.
Temperature.	Best growth at 30° to 37° C. Somewhat slower at room temperature.
Rapidity of growth.	Quite rapidly.
Spore formation.	Spore formation has not yet been observed, but it possesses a remarkable power of resistance against destructive influences.
Aerobiosis.	Exists quite a long time without air (Rosenbach); does not produce fetid putrefaction.
Gas production.	Does not form gas.
Gelatin reaction.	Liquefies gelatin.
Aniline reaction.	Is colored well by Gram's method. Color product is orange-yellow.
Pathogenesis.	Effect upon animals differs according to the mode of application. Subcutaneous inoculation is without effect in mice, guinea-pigs, and rabbits, with the exception of localized abscess at the point of inoculation in the two latter animals. Injected into the abdominal cavity, they are borne for a short time; into the vessels, they set up inflammation of the joints and kidneys; later, they attack the valves of the heart, and set up a typical endocarditis ulcerosa. (Orth, Wyssokowitsch, Ribbert.)

84. Micrococcus of osteomyelitis. *Becker.*

D. med. Wochenschr. 1883. No. 46.

Place found.	In the pus of osteomyelitic foci.
Form and arrangement.	Micrococci, single and arranged in heaps.
Motility.	Non-motile.
Growth.	
Gelatin.	*Plate Culture.*—After three to twenty-four hours fine dots of a yellow or orange color occur. *Tube Culture.*—At room temperature whitish lines develop along the puncture on the third day. Liquefaction occurs from the surface, and at the point where the liquefied gelatin is in contact with the still solid media an intense orange sediment is found. Cultures exposed to the air possess an odor of sour paste.
Potatoes. Blood-serum.	The growth is facilitated by a temperature of 36° C. It appears after twenty-four hours as an orange-colored coating. At 30° C. the growth occurs as a whitish cloudiness along the stroke, which by degrees assumes an orange color.
Temperature.	Best growth at 30° C.
Rapidity of growth.	Grows rapidly.
Formation of spores.	
Aerobiosis.	Facultative aerobic.
Gas production.	
Gelatin reaction.	Liquefying.
Aniline reaction.	Aqueous solution of methyl-blue forms the best coloring agent.
Pathogenesis.	Death of rabbits occurs in twelve to fourteen days after an intravenous injection. If the bones are bruised or fractured, suppuration occurs at the points so treated. The cocci are found in the blood and in the pus. Nos. 83 and 84 are probably identical.

85. Staphylococcus pyogenes citreus. *Passet.*

Place found.	In pus. In air. (Pawlowsky.)
Form and arrangement.	
Motility.	
Growth.	
Gelatin.	
Agar-agar.	
Potatoes.	
Blood-serum.	
Temperature.	
Rapidity of growth.	
Formation of spores.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	
Aniline reaction.	
Pathogenesis.	

Morphologically as well as physiologically entirely like No. 83, except that it produces lemon-yellow pigment.

86. Staphylococcus pyogenes albus. *Rosenbach.*

Place found.	In pus.
Form and arrangement.	Like No. 83, except that it is without the formation of pigment.
Motility.	
Growth.	
Gelatin.	
Agar agar.	
Potatoes.	
Blood-serum.	
Temperature.	
Rapidity of growth.	
Spore formation.	
Aerobiosis.	Still retains vitality after three and a half years, and this, too, under anaerobic conditions.
Gas production.	
Gelatin reaction.	Liquefying.
Aniline reaction.	
Pathogenesis.	

87. Micrococcus pyogenes tenuis. *Rosenbach.*

Place found.	In extensive but rather benign phlegmons. (Infrequent.)
Form and arrangement.	Cocci, somewhat larger than the staphylococcus, and without any especial arrangement.
Motility.	
Growth.	
Agar-agar.	*Plate Culture.*—A surface growth somewhat hyaline in character. *Tube Culture.*—Growth occurs along the inoculation stroke as a very thin, vitreous layer, resembling a streak of laquer.
Potatoe.	
Blood-serum.	
Temperature.	
Rapidity of growth.	Grows slowly.
Spore formation.	
Acrobiosis.	
Gas production.	
Gelatin reaction.	Not studied.
Aniline reaction.	The larger individuals color more intense at the poles.
Pathogenesis.	Animals inoculated give negative results.

88. Staphylococcus cereus albus. *Passet.*

Place found.	In pus.
Form and arrangement.	Cocci of irregular size, arranged in heaps, more rarely in chains.
Motility.	Non-motile.
Growth.	
Gelatin.	*Plate Culture.*—Small white dots, which spread upon the surface until they reach 1 to 2 mm. in diameter. *Tube Culture.*—Surface growth of a grayish-white color, resembling drops of stearin, or wax, the edges of which are thickened and somewhat irregular. In stab culture they develop as a grayish-white line composed of minute colonies. (Staubchen.)
Potatoes.	Grayish-white coating of medium depth.
Blood-serum.	Along the stroke a pale, grayish-white line develops.
Temperature.	Grows at room temperature.
Rapidity of growth.	Rather rapid growth.
Formation of spores.	Not known.
Aerobiosis.	
Gas production.	Not observed.
Gelatin reaction.	Not liquefying.
Aniline reaction.	
Pathogenesis.	

89. Staphylococcus cereus flavus. *Passet.*

Place found.	In pus.
Form and arrangement.	
Motility.	
Growth.	
Gelatin.	
Agar-agar.	
Potatoes.	
Blood-serum.	
Temperature.	
Rapidity of growth.	
Spore formation	
Acrobiosis.	
Gas production.	
Gelatin reaction.	
Aniline reaction.	
Pathogenesis.	

Like No. 88, except that its pigment is yellow. (Darker than No. 85.)

90. Staphylococcus salivarius pyogenes. *Biondi.*

Zeitschr. f. Hygiene, Bd. II, S. 227.

Place found.	In an abscess occurring in a guinea-pig inoculated with the saliva of a patient suffering from angina scarlatinosa.
Form and arrangement.	Round cocci, smaller than most known at present, 0.3 to 0.5 m. in diameter. In the pus they were arranged in the form of grape clusters, differing in this from No. 83 to No. 86.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Under and on the surface round, sharply defined, opalescent colonies occur, of a whitish color; they slowly liquefy the geletin surrounding them. *Tube Culture.*—At 12° to 14° C. growth begins at the surface, spreads along the stroke, and in eight days has occasioned the "funnel-shaped" depression. At the apex small, white, shining granules are deposited, and on the surface of the liquefied gelatin a whitish film is formed, of a slimy, tenacious character.
Agar-agar.	*Tube Culture.*—At culture temperature a thick and broad streak results, about 1 mm. broad, extending along the inoculation stroke, of a golden-yellow color.
Bouillon.	If kept in the incubator a cloudiness occurs after two hours, which later settles to the bottom as a compact sediment.
Temperature.	Exhibits a great resistance to low temperatures and to the drying methods. Kept for three months at 8° to 9° C., cultures retained their power of procreation and virulence, as also when kept at 40° C.
Rapidity of growth.	Grows very rapidly in agar, slowly in gelatin.
Spore formation.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Liquefies gradually.
Aniline reaction.	Colors the best by Gram's method.
Pathogenesis.	Local, circumscribed suppuration in mice, guinea-pigs, dogs, rabbits, after subcutaneous inoculation.

91. Coccus salivarius septicus. *Biondi.*

Zeitschr. f. Hygiene, Bd. II, S. 217.

Place found.	In saliva of a patient suffering from severe puerperal septicæmia.
Form and arrangement.	Round cocci, possessing, in stage of active division, narrow, median fissures. They occasionally assume a slightly oval form.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Beneath the surface, colonies of a whitish-gray color, sometimes bordering on black. *Tube Culture.*—The growth consists of dot-like, whitish colonies, of a markedly granular appearance.
Agar-agar.	*Tube Culture.*—A rich growth on the surface.
Potatoes.	Very slight growth.
Blood serum.	
Temperature.	Active growth at incubator temperature. Grows also at 18° to 20° C.
Rapidity of growth.	Grows quite rapidly.
Spore formation.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Colors well with all aniline colors; also by Gram's method.
Pathogenesis.	Pathogenic to mice, guinea-pigs, and rabbits, which are killed in from four to six days. Large numbers of cocci are found heaped in the organs and in the blood. Inflammatory symptoms are absent.

92. Bacillus saprogenes I. *Rosenbach.*

Mikroorganismen bei Wundinfektionskrankheiten. Wiesbaden, 1884.

Place found.	In white thrombi of the mucous membrane of the fauces.
Form and arrangement.	Rather larger bacilli; at the ends of individual bacilli large spores are visible.
Motility.	
Growth.	
Gelatin.	
Agar-agar.	*Tube Culture.*—A yellowish gray, opaque streak occurs along the puncture, having a height of 1 mm.; it has a pap-like, sticky consistency. After a time the edges become wavy, and in from four to five weeks it emits an odor resembling that of decomposed kitchen garbage.
Potatoes.	
Blood-serum.	The growth emits an intense putrid odor.
Temperature.	
Rapidity of growth.	Grows slowly.
Spore formation.	Spores are terminal.
Aerobiosis.	Aerobic. With admission of air the intense, putrescent odor is developed. Under anaerobic conditions the growth is very slight.
Gas production.	
Gelatin reaction.	
Aniline reaction.	
Pathogenesis.	Injected into the knee-joints of rabbits, they gave negative results.

93. Bacillus saprogenes II. *Rosenbach.*

Place found.	In the secretion of bromidrosis pedis.
Form and arrangement.	Bacilli, somewhat smaller than No. 92.
Motility.	
Growth.	
Agar-agar.	*Tube Culture.*—After twenty to twenty-four hours numerous fine, transparent drops develop along the stroke, which increase in size and coalesce. At first they are white, later gray and mucilaginous. The culture yields the characteristic odor of sweating feet.
Potatoes.	
Blood-serum.	
Temperature.	
Rapidity of growth.	A very rapid surface growth.
Formation of spores.	
Aerobiosis.	Aerobic. Facultatively anaerobic. Under the latter condition they emit a disgusting, putrid odor; under the former the odor is less disagreeable.
Gas production.	Emits odor resembling that from sweating feet.
Gelatin reaction.	No growth.
Aniline reaction.	
Pathogenesis.	Has the power of invasion, and possesses pyogenic qualities.

94. Bacillus saprogenes III. *Rosenbach.*

Place found.	In septic, gangrenous pus.
Form and arrangement.	Short, thick staves, with rounded corners.
Motility.	
Growth.	
Agar-agar.	*Tube Culture.*—At room temperature an ash-gray, almost liquid growth develops after eight days, 3 mm. in diameter, and having a wavy appearance.
Potatoes.	
Blood-serum.	
Temperature.	
Rapidity of growth.	Growth of medium rapidity. (Maximum in eight days.)
Spore formation.	
Aerobiosis.	Under anaerobic conditions albumen is quickly decomposed; under aerobic conditions the decomposition is slower. Cultures on all media, excepting milk, emit a putrid odor.
Gas production.	Cultures emit a foul odor.
Gelatin reaction.	No growth.
Aniline reaction.	
Pathogenesis.	A yellowish-green infiltration, accompanied by an extensive injection and a slightly putrid odor, follows subcutaneous or direct inoculation into the joints of rabbits.

95. Bacillus pyogenes fœtidus. *Passet.*

Ætiologie der eiterigen Phlegmone des Menschen. Berlin, 1885.

Place found.	In an abscess of the anus.
Form and arrangement.	Short staves with rounded ends, often arranged in chains of two or more.
Motility.	Slightly motile.
Growth.	
Gelatin.	*Plate Culture.*—In twenty-four hours little white dots develop. Those on the surface reach the size of a three-cent piece, or are confluent. In their centre they are thick and white, the periphery thinner and gray. *Tube Culture.*—In twenty-four hours a superficial, delicate, grayish-white veil develops, with somewhat thicker, irregular edges. Along the puncture itself a collection of dots develop, which, at first quite delicate, become larger. Older cultures cause a turbidity of the upper portion of the gelatin.
Potatoes.	A shining, luxuriant, light-brown growth results.
Blood-serum.	Along the stroke a medium thick, grayish-white streak.
Temperature.	Grows at room temperature.
Rapidity of growth.	Grows rapidly
Spore formation.	Probably forms spores.
Aerobiosis.	
Gas production.	Develops always a disagreeable stench.
Gelatin reaction.	Non-liquefying.
Aniline reaction.	
Pathogenesis.	Injected subcutaneously and into the abdominal cavity of mice and guinea-pigs, they cause death in twenty-four hours. Numerous bacilli are found in the blood, but not at the point of inoculation nor in the organs.

96. Streptococcus pyogenes. *Rosenbach, Passet.*

Place found.	In progressive erysipeloid suppuration.
Form and arrangement.	Cocci arranged in chains, consisting of as many as thirty members; often in pairs.
Motility.	
Growth.	
Gelatin.	*On Plates.*—Round, finely granular, little dots along the inoculation stroke. The cultures are thickest at the middle, and have a brownish color. By degrees the growth becomes thicker, punctate, wavy, heaped up, and later terraced. *Tube Culture.*—Delicate halo around the puncture, which is finely granular; or, it consists of larger and smaller dots, with larger ones below.
Agar-agar.	*Tube Culture.*—At 35° to 37° C. a continuous, grayish-white line develops, accompanied by little dots. No halo appears on the surface.
Potatoes.	They do not multiply on potatoes, but individual cocci increase in size, so that under the microscope we may see larger and smaller cocci.
Blood-serum.	*Tube Culture.*—As a thin, continuous line.
Temperature.	Grows best at 35° to 37° C. Room temperature is less favorable.
Rapidity of growth.	Growth is slow. In tube cultures it is only 2 to 3 mm. in breadth in two to three weeks. In four months the culture is nearly lifeless.
Spore formation.	
Aerobiosis.	Not especially affected by aerobic conditions. In vacuo it decomposes albumen.
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Remains stained after Gram's method.
Pathogenesis.	Produces a progressive erysipeloid suppuration.

97. Streptococcus erysipelatis. *Fehleisen.*

Die Ætiologie des Erysipels. Berlin, 1883.

Place found.	In the cutaneous lymph-channels of erysipelas.
Form and arrangement.	Very small cocci, arranged in pairs or in long chains. The latter arrangement is especially common in cultivations in bouillon.
Motility.	
Growth.	
Gelatin.	*On Plates.*—Colonies are small, round, and finely granular. *Tube Culture.*—After twenty-four hours a growth resembling dust, interspread with dotlets, develops along the inoculation stab, which later coalesces into a homogeneous, opaque, white line. On the surface the growth is absent or so small as to be scarcely visible.
Agar-agar.	*Plate Culture.*—At incubator temperature a rather rapid development occurs as a delicate, transparent, gray-colored, drop-like growth, which does not spread but very little. *Tube Culture.*—Growth sparing and situated on the entire surface in colonies so small as to be almost invisible.
Potatoes.	No growth.
Blood-serum.	At 37° C. a pretty, white tuft develops, which is easily removable from the surface.
Temperature.	Best results at 30° to 37° C.
Rapidity of growth.	Grows slowly.
Spore formation.	Forms no spores; we must, however, on clinical ground assume that there is some durable form.
Aerobiosis.	Facultative aerobic.
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Retains color after Gram's method.
Pathogenesis.	It causes in rabbits a sharply limited hyperæmia, which spreads and is unaccompanied by suppuration. The germ is constant in the lymph-spaces of the skin in erysipelas. Transferred from one person to another, it proved successful several times, a typical erysipelas occurring each time. There is a probability that Nos. 96 and 97 are identical. Their cultures are not to be distinguished one from another. Yet, we must admit this difference: that while the living tissue furnished a most favorable nourishing media for the streptococcus, for the erysipelas coccus it is very poor.

98. Streptococcus pyogenes malignus. *Flügge.*

Flügge, Mikroorganismen. Leipzig, 1886. S. 153.

Place found.	From necrotic foci of leucæmic spleen.
Form and arrangement.	Not to be distinguished from Nos. 96 and 97.
Motility.	
Growth.	
Gelatin.	Like Nos. 96 and 97, but the colonies appear to be smaller.
Agar-agar.	
Potatoes.	
Blood-serum.	
Temperature.	
Rapidity of growth.	Grows slower than Nos. 96 and 97.
Formation of spores.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	
Pathogenesis.	Inoculated mice die in from three to five days. Post-mortem shows suppurating area at point of inoculation, together with the cocci in the blood and scattered through the parenchymatous organs. Rabbits subjected to intra-venous inoculation die in four days, and at the point of inoculation give the same results as stated in Nos. 96 and 97. Section the same as in mice.

99. Streptococcus septicus. *Nicolaier, Guarneri.*

Flügge, Mikroorganismen. Leipzig, 1886. S. 154.

Place found.	In soiled earth.
Form and arrangement.	Similar to the other streptococci, but do not possess so distinct a tendency to form chains under all conditions. In the tissues diplococci forms are most frequently found.
Motility.	
Growth.	
Gelatin.	*On Plates.*—The colonies resemble those of the other streptococci, —small, white dots.
Potatoes.	
Blood-serum.	
Temperature.	
Rapidity of growth.	Grows even slower than No. 98.
Formation of spores.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	
Pathogenesis.	Without exception inoculated mice die after forty-eight to seventy-two hours. During the last twenty-four hours a distinct paresis of both sensation and motion in the hind extremities ensues. In the blood and organs large numbers of diplococci are found. In rabbits inoculated in the ear-vein, the first change occurring is the local hyperæmia, and death ensues in two to three days. In the organs immense numbers of cocci are found, which by degrees accumulate to such an extent as to form thrombi, and thus to necrotic foci. This tendency to thrombi and necrotic foci, according to Oberdick, causes the transfer of the streptococci to the fœtus.

100. Streptococcus septo-pyæmicus. *Biondi.*

Zeitschr. f. Hygiene, Bd. II, S. 225.

Place found.	From the saliva of individuals, of whom one was afflicted with angina phlegmonosa and two with primary laryngeal erysipelas.
Form and arrangement.	Perfectly round cocci, 0.7 to 0.8 m. in diameter, arranged in pairs and chains of different sizes.
Motility.	
Growth.	
Gelatin.	*On Plates.*—After forty-eight hours at cultivation temperature, ovoid, yellowish-gray colonies with rough edges appear. *Tube Culture.*—In the form of finely granular, white dots, which are somewhat coarser at the periphery than in the middle.
Agar-agar.	*On Plates.*—Same as on gelatin, more rapid in growth.
Potatoes.	In small, plateau-like, dirty-white colonies, which never reach large dimensions.
Blood-serum.	Same as on potatoes.
Temperature.	
Rapidity of growth.	
Spore formation.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	
Pathogenesis.	Not invariably pathogenic to guinea-pigs, mice, and rabbits; injected into the auricular vein of the latter, they cause a true erysipeloid inflammation. Probably identical with No. 97.

101. Streptococcus articulorum. *Loeffler.*

Mitth. a. d. Kais. Ges.-Amt., Bd. II, S. 451. Flügge, Mikroorg., S. 153.

Place found.	In different types of diphtheria, in and on the diseased mucous membrane.
Form and arrangement.	Cocci forming chains containing as many as one hundred members. In individuals there is a slight intimation of a median fissure.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Forms small, limpid, light-gray drops. Under low power their rims are resolved into small, curled lines (coccus chains).
Potatoes.	Growth very slow. After eight days they are only microscopically visible.
Blood-serum.	As a thin, pale, glimmering coating, similar to the layer of cholesterin that forms on serum. The best nourishing media is serum with one-fourth volume neutral meat infusion-peptone-sodium-chloride solution. At 37° C. a distinct, grayish-white layer appears after one to two days.
Temperature.	Grows at room or incubator temperature.
Rapidity of growth.	Grows slowly.
Formation of spores.	Not known.
Aerobiosis.	
Gas production.	Not observed.
Gelatin reaction.	Non-liquefying.
Aniline reaction.	
Pathogenesis.	Pathogenic to mice; guinea-pigs are immune. Intra-venous injections in rabbits produce at first typical joint-disease, from which death slowly results in the majority of cases.

102. Bacillus pyocyaneus. *Gessard.*

De la pyocyanie et de son microbe. 1882.

Place found.	In green pus.
Form and arrangement.	Short, fine staves, sometimes easily mistaken for cocci.
Motility.	Actively motile.
Growth.	
Gelatin.	*On Plates.*—In two to three days the whole plate has assumed a light-green color; the surface colonies liquefy in the "funnel-form." Under low power, the deeper colonies appear round, yellowish, and transparent, with refractive, granular rims. *Tube Culture.*—Stab inoculation: After twenty-four hours the gelatin is superficially liquefied and depressed (funnel-form), while the surface of the firm gelatin assumes a fluorescence. Liquefaction becomes separated horizontally from the gelatin, and the fluorescence spreads through the entire extent of the media. Stroke inoculation: Moist, greenish-white coating. The entire media assumes a diffuse fluorescence.
Agar-agar.	*Tube Culture.*—Moist, greenish-white layer develops, after which the entire contents of the tube assumes a pretty, diffuse fluorescence.
Potatoes.	Dry, rust-brown colonies develop, which are confined to the inoculation stroke. When moistened with ammonia they assume a green color; with acids, a red color.
Milk.	They produce on the surface yellowish-gray flecks, separate casein, and peptonize it with production of ammonia.
Temperature.	Grows well at room temperature.
Rapidity of growth.	Grows rapidly.
Spore formation.	Not observed.
Aerobiosis.	Does not grow under mica plate.
Gas production.	
Gelatin reaction.	Liquefying.
Aniline reaction.	Coloring matter, a blue body called pyocyanin. (Fordos and Gessard).
Pathogenesis.	Injected into the abdominal cavity, caused the death of guinea-pigs (Koch). Rabbits live after intra-venous injection.

103. Micrococcus botryogenus. *Johne, Rabe.*

Zeitschr. f. Thiermed. u. Path., Bd. XII, S. 137.

Place found.	Connective-tissue tumors of the perimysium and the subcutis, the spermatic cord, and in the retroperitoneal tissue of the pelvic region of the horse.
Form and arrangement.	Micrococci, 1 to 1.5 m. in diameter, arranged in the form of wavy chains.
Motility.	Non-motile.
Growth.	
Gelatin.	*On Plates.*—Circular, sharply-defined colonies, at first silver-gray and later of a more yellowish-gray color and metalic lustre. In time the plate has the appearance of having been dusted over with dried blood, and is distinguished by a peculiar fruity, aromatic, refreshing odor, reminding one of strawberries. *Tube Culture.*—At first a pale, grayish-white line develops, around which the gelatin is liquefied. Later the color is changed to a milk-white, and at the upper end of the stroke a characteristic bubble appears.
Agar-agar.	*Tube Culture.*—A hardly noticeable growth.
Potatoes. Blood-serum.	In the form of yellowish, frost-like covering, with an odor the same as from the plate cultures. They are the most favorable nourishing media.
Temperature.	Growth at room temperature.
Rapidity of growth.	Liquefaction is hardly discernible.
Spore formation.	
Acrobiosis.	
Gas production.	On gelatin or potatoes the cultures generate a characteristic odor, refreshing in its effect, resembling that of strawberries.
Gelatin reaction.	Scarcely discernible liquefaction.
Aniline reaction.	
Pathogenesis.	Guinea-pigs are killed with symptoms of septicæmia. In sheep and goats they cause a severe inflammatory process to spread from the point of inoculation. Mice seem to be immune. In horses an inflammatory œdema is at first set up, followed in four to six weeks by connective-tissue nodules which sometimes break down. They contain large numbers of micrococci.

104. Micrococci of progressive lymphoma of animals. *Manfredi.*

Fortschritte der Med. 1886. S. 713.

Place found.	In the sputum of pneumonia subsequent to morbilli.
Form and arrangement.	Oblong micrococci, with rounded and blunt ends, most frequently single; occasionally arranged as diplococci. Their size averages about 0.4 to 0.6 m.
Motility.	
Growth.	
Gelatin.	*On Plates*—Colonies in the form of thin, transparent plaques; in direct light, of a bluish color; with reflected light, of a pale, gray color and slightly mottled surfaces. *Tube Culture.*—Grows slowly along the stroke as a delicate, grayish-yellow line, which remains stationary when formed. The surface end is crowned with a plaque.
Potatoes.	Growth slight. At 37° C., a very thin, delicate, moist, slightly granular layer is formed, which is rather lustrous and of a yellowish color.
Blood-serum.	In the form of delicate *patinæ*, with irregular borders, lustrous surface, and colored a yellowish green.
Temperature.	Favorable temperature ranges from 18° to 37° C. At 46° to 48° C., growth begins to cease, and at 48° to 60° C. it has ceased entirely, as well as its power of rejuvenation.
Rapidity of growth.	Grows rapidly on the surface of gelatin.
Spore formation.	
Aerobiosis.	Aerobic conditions are more favorable.
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	A characteristic staining has not as yet been found. They color well with Gram's method.
Pathogenesis.	Inoculated animals—dogs, rabbits, guinea-pigs, domestic mice—die on an average in seven to twelve days, and their parenchymatous organs exhibit an enormous increase in size, and within them gray and yellowish-gray nodules which belong to the granuloma. These undergo caseation, contain the specific micro-organisms, and are themselves infectious.

105. Bacillus of rhinoscleroma. *v. Frisch.*

v. Frisch, Wien. Med. Wochenschr. 1882. No. 32. Paltauf u. v. Eiselsberg, Fortschr. d. Med. 1886. No. 19.

Place found.	In the connective-tissue juice of rhinoscleroma.
Form and arrangement.	Short bacilli, the length of which is two to three times greater than their breadth, the ends of which are rounded; or they occur as oval cocci, usually united in pairs, which can grow to bacilli and seemingly into threads.
Motility.	Non-motile.
Growth.	The growth is similar to Friedlaender's pneumo-bacillus.
Gelatin.	*On Plates.*—On the second or third day, sharply circumscribed, whitish-yellow, round colonies appear. Under low power they are distinctly granular, and the superficial colonies are of a bright-gray color. *Tube Culture.*—Typical "nail" culture, the head of which is white, moist, and lustrous, and of a tough, mucilaginous consistency.
Agar-agar.	*On Plates.*—At 36° to 38° C., after twelve hours, the plate appears as though milk had been poured upon it and partially curdled.
Potatoes.	As white or slightly yellowish growth, which later spreads over the entire surface. A gas is often formed.
Blood-serum.	As a whitish layer, which sinks in the water of condensation.
Temperature.	Grows best between 36° to 38° C.
Rapidity of growth.	Grows rather rapidly.
Formation of spores.	
Aerobiosis.	Facultative aerobic.
Gas production.	Often develops a gas on potato culture.
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Colors well with Loeffler's methylene-blue solution. By staining with aniline, gentian violet, and after-treated with acetic acid or with Ziehl's carbolic-fuchsin solution the bacilli appear to be surrounded by a capsule. They are partially decolorized by Gram's method.
Pathogenesis.	The bacilli, inoculated into mice, guinea-pigs, etc., cause inflammation of pleura, abscesses in the subcutaneous and muscular tissue, and death of the animal. Their virulence is not so great as that of Friedlaender's pneumo-bacillus.

106. Bacillus mallei. *Loeffler, Schütz.*

D. Med. Wochenschr. 1882. No. 52. Weichselbaum, Wien. Med. Wochenschr. 1885. Nos. 21–24.

Place found.	In the nodules occurring in glanders.
Form and arrangement.	Staves the size of tubercle bacilli. They exceed them in breadth, however, being 2 to 5 m. long and 0.5 to 1.1 m. broad.
Motility.	Very motile.
Growth.	
Gelatin.	*Tube Culture.*—At 37° C., whitish growth occurs, composed of chains and threads coiled and twisted in many directions, which penetrate the gelatin. *On Plates.*—At 37° C., bright-yellow or lustrous white colonies.
Agar-agar.	*Tube Culture.*—Drop-like, soft, grayish-white colonies. (Weichselbaum.)
Potatoes.	At 37° to 38° C., on mashed potatoes, a brown, paste-like mass appears, similar in appearance to that of Koch's comma bacillus. At room temperature there occurs only a slight growth.
Blood-serum.	At 37° C., on the third day, growth is apparent as numerous small, lustrous, drop-like colonies, scattered over the surface.
Glycerin agar-agar.	More luxuriant growth than on any other media, even at room temperature. They form a pale-white, transparent stria (7 to 8 mm.) along the puncture. (Kranzfeld.)
Temperature.	Growth best at incubating temperature,—between 30° and 40° C., and not over 43° C., nor under 25° C.
Rapidity of growth.	Grows slowly.
Spore formation.	Forms spores (Weichselbaum). They can retain their life-power in a dry condition for nearly three months.
Aerobiosis.	
Gas production.	Not observed
Gelatin reaction.	Slight growth.
Aniline reaction.	Loeffler's concentrated alcoholic solution of methylene blue is the best stain, and after-treated with oxalic-sulphuric acid, alcohol, cedar-oil. Gram's method is not applicable.
Pathogenesis.	Pure cultures injected into horses, rabbits, guinea-pigs, and field-mice produce typical glanders. Guinea pigs die in about six to eight weeks, field-mice in a few days. Bacilli are found abundantly in the newly-formed nodules and abscesses, as well as in the blood and urine. White mice are immune. As animals sensitive to the inoculation, Kranzfeld recommends the *Spermophilus guttatus;* Kitt, the forest mouse,—*Mus sylvaticus.* In the latter, however, a longer incubation stage is required.

107. Bacillus tuberculosis. *Koch.*

Mitth. a. d. Kais. Ges.-Amt., Bd. I.

Place found.	In all the products of human and animal tuberculosis.
Form and arrangement.	Very thin staves, 2 to 5 m. long,—¼ to ½ the diameter of a red blood-corpuscle,—nearly always beaded or spotted. Their ends are rounded
Motility.	Non-motile.
Growth.	
Gelatin.	No growth.
Glycerin agar-agar.	*Tube Culture.*—A luxuriant growth in the form of isolated, folded or wrinkled, smutty-white scales. (Nocard Roux.)
Potatoes.	At cultivation temperature, whitish, smooth colonies form about the twelfth to twentieth day; these are easily loosened from the surface of the media. The potato must be hermetically enclosed in test-tubes. (Pawlowski.)
Blood-serum.	Colonies take the appearance of pale, white, lustrous dots, comparable to small, dry scales. Numerous, and closely-arranged, they form a grayish-white layer on the surface of the water of condensation, in which it does not sink, but remains floating on its surface. Under a low power, dainty reticulations appear, consisting entirely of bacilli.
Temperature.	Optimum, 37.5° C. Under 30° and above 42° C. they do not develop.
Rapidity of growth.	Grows very slowly. Only visible after ten to fifteen days. In four weeks the maximum development is reached.
Spore formation.	Spore formation occurs with an animal body. The exact mode of spore formation is not as yet definitely settled.
Aerobiosis.	
Gas production.	Not observed.
Gelatin reaction.	No growth.
Aniline reaction.	*Ehrlich:* Twenty-four hours in aniline-water solution of fuchsin or gentian violet, a few seconds in nitric acid (1 to 4): then rinse in 60-per-cent. alcohol; after, stain with a weak, aqueous solution of methylene blue or Bismarck brown. *Rindfleisch:* (especially useful in cover-glass preparations of sputum) The same as above, except that the stains are heated over a small alcohol-flame until bubbles arise. *Gabett-Ernst:* See Appendix.
Pathogenesis.	Injection of tubercular products and pure cultures cause tuberculosis in animals. The bacilli are found in the nodules and especially in the giant-cells, in large numbers or singly. Found in the blood of those suffering from miliary tuberculosis. Field-mice, rabbits, and cats are inoculable. White mice, rats, and dogs are immune.

108. Lepra bacillus. *Armauer Hansen.*

Virchow's Archiv, Bd. LXXIX u. XC. Neisser, Ziemssen's Hdbch., Bd. XIV. Virchow's Archiv, Bd. CIII, S. 355.

Place found.	In all the specific products occurring in the leprous process.
Form and arrangement.	Small, slim staves, about half to two-thirds the diameter of a red blood-corpuscle, with ends that are occasionally pointed. According to Bardoni they are club-shaped at the ends.
Motility.	Non-motile.
Growth.	The addition of glycerin renders all media more suitable.
Gelatin.	*Plate Culture.*—The first generation does not develop upon gelatin. Later, irregular, round colonies develop, both beneath and upon the surface, at 20° to 25° C.
Agar-agar.	*Plate Culture.*—At 35° to 37° C., gray, roundish flakes develop. Under low power the centres appear dense, the rims irregular and jagged, composed of a fine, irregularly-arranged, flocculent reticulum. *Tube Culture.*—Whitish-gray, roundish colonies, with prominent centres and jagged edges.
Blood-serum.	On desiccated blood-serum, or on boiled chicken or duck-eggs, a small nodule appears, about the size of a millet-seed, which in three weeks is increased to double its original size, in the form of a narrow zone. (Neisser.) At 35° to 37° C. band-like colonies develop along the inoculation stroke, the edges of which are irregular. Their color is light-yellow (waxy). The serum is not liquefied.
Potatoes.	No growth.
Temperature.	Best growth at 37° to 38° C. They grow, however, at 20° C.
Rapidity of growth.	Grows very slowly.
Spore formation.	*Endosporulation.*—Two to three spores in each cell, having the appearance of oval vacuoles.
Aerobiosis.	
Gas production.	
Gelatin reaction.	Not liquefied.
Aniline reaction.	They have the same stain reaction as the bacillus tuberculosis, but are colored more quickly, and they also accept the ordinary nuclear stains, as well as Gram's. They do not stain in Loeffler's aqueous, alkaline, methyl-blue solution, thus differing from the tubercle bacillus. (Neisser.)
Pathogenesis.	Local leprous manifestations may be produced on animals by vaccination (Damsch and Vossius), but no general symptoms of leprosy.

109. Tetanus bacillus. *Nicolaier, Kitasato.*

Nicolaier, D. med. Wochenschr. 1884. No. 52. Kitasato, Zeitschrift f. Hyg., Bd. VII, S. 225.

Place found.	Earth, and in the secretions from the wounds of those afflicted with tetanus.
Form and arrangement.	Fine bacilli, somewhat longer, hardly finer than Koch's mouse-septicæmia bacillus. They form threads, in irregular masses, which have a setaceous appearance in the sporulation stage.
Motility.	Possibly motile, but very slightly.
Growth.	Pure cultures are to be obtained from pus with certainty by first heating the mixed cultures one half to one hour for several days in a water-bath at 80° C., after which the plate-method is employed in an atmosphere of hydrogen. Von Kitt employs a method by which the pus is greatly attenuated.
Gelatin (containing 1.5–2 per cent. of grape-sugar).	*Plate Culture.*—After four to five days at 20° to 25° C., or in a week at 18° to 20° C., colonies resembling those of the hay bacillus develop. They are discreet, the centres are dense; they develop regularly in all directions in the form of a nimbus. Laterally, liquefaction slowly occurs and gas is formed. *Higher Culture.*—Growth occurs along the stroke and under the surface, as a radiating cloudiness, often possessing aculeated, radiating apophyses. Later, liquefaction and the production of gas.
Agar-agar.	*Tube Culture.*—Most rapid growth if 1 to 2 per cent. of grape-sugar be added.
Bouillon.	As a dense cloudiness.
Blood-serum.	In one to three days, at 34° to 38° C., crescentic, isolated colonies, lying in shallow depressions, occur, which, later, become confluent and give an undulated appearance to the surface of the gelatin, from which the condensation water arises. After six to ten days, transverse division of the solid serum and softening of the bases of the undulations occur.
Temperature.	Grows best at 36° to 38° C. Under 16° C., no growth.
Rapidity of growth.	Grows rather slow.
Spore formation.	Spore formation is characteristic. At 37° C., the bacilli assume a setaceous appearance after thirty hours, and round terminal spores are formed, which possess a rather marked resistance to destructive agencies. One hour's exposure to 80° C., in moisture, is not destructive to them, though six minutes in steam, at 100° C., are fatal. Dried spore-containing pus remains virulent after sixteen months. (Kitt.)
Aerobiosis.	Obligate anaerobic. Grows only when air is excluded; well under hydrogen; not under carbonic acid.
Gas production.	Cultures give off a disagreeable odor.
Gelatin reaction.	Liquefies.
Aniline reaction.	Colors equally well with aniline dyes, and by Gram's method.
Pathogenesis.	Domestic mice, inoculated at the roots of the tails with earth that contains the bacilli, remain unaffected for one and a half to two and a half days. Then tetanic spasms begin in one hind-leg, followed by the other. The fore-legs then become fixed, then follow reflex tetanic spasms, and death ensues in three to five days later. Rabbits and guinea-pigs are especially susceptible. Inoculations with cultures on blood-serum were equally successful. *Section.*—No suppuration at the point of inoculation; no bacilli or spores to be found in the organs or blood. They seem to disappear very rapidly from the organism, though the tetanic phenomena remain.

110. Micrococcus tetragenus. *Koch, Gaffky.*

Mitth. a. d. Kais. Ges.-Amt., Bd. II, S. 42. v. Langenbeck's Archiv, Bd. XXVIII, Heft 3, S. 500.

Place found.	In the caverns and sputum of pulmonary phthisis. (Biondi.)
Form and arrangement.	Small cocci, lying together in twos and fours. Those procured from the body are found arranged in fours, which are inclosed in clear capsules that do not stain.
Motility.	Non-motile.
Growth.	
Gelatin.	*On Plates.*—Small, white, dot-like colonies, which, under a low power, are finely granular, and possess a characteristic, vitreous lustre. *Tube Culture.*—Growth does not occur along the entire inoculation stroke; round, sharply-defined, milk-white or yellowish, lenticular colonies occur in isolated patches throughout the gelatin.
Agar-agar.	*Tube Culture.*—Growth occurs along the stroke. The colonies are circumscribed, round, and white.
Potatoes.	Forms a thick, slimy coating, which may be removed in long shreds.
Blood serum.	A white, moist circuitous streak.
Temperature.	Grows at warm temperature.
Rapidity of growth.	Grows slowly.
Formation of spores.	Not known.
Aerobiosis.	Facultative aerobic.
Gas production.	Not observed.
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Accepts the common nuclear stains as well as Gram's double stain.
Pathogenesis.	Mice and guinea-pigs inoculated with the micrococcus die in from three to ten days. In the blood and organs large numbers of the organism are found. Field mice and rabbits are not susceptible.

III. Typhus baciilus. *Eberth, Gaffky.*

Mitth. a. d. Kais. Ges.-Amt., Bd. II.

Place found.	In blood, urine, fæces, and tissues of typhoid patients.
Form and arrangement.	Bacilli three times as long as they are broad, accompanied by short staves, with rounded ends. They can form threads, and often have a filiform appearance. They do not stain so readily with the aniline colors as most similar organisms.
Motility.	Very motile.
Growth.	
Gelatin.	*On Plates.*—The entire surface is occupied by grayish-white colonies, which have scolloped edges. Under a low power braids resembling glass-wool, and of a brownish color, are visible. *Tube Culture.*—Slight growth along the puncture; more on the surface, in the form of a grayish-white layer, with jagged edges.
Agar-agar.	*Tube Culture.*—Growth is superficial and of a whitish color.
Potato-gelatin.	*Tube Culture.*—Similar to gelatin, with this difference, however, that the colonies here lack the yellowish tint, and possess a characteristically shining and transparent appearance upon the surface. (Hotz.)
Potatoes.	After forty-eight hours the growth is still imperceptible, but the surface of the potato has a peculiar, moist appearance, which, if touched with the platinum needle, gives the impression of having an adherent skin. Examined microscopically, it is found to be composed of long spores containing threads of typhus bacillus. The growth becomes visible when grown on mashed potato, treated with alcohol, and kept at incubation temperature. It then has a dirty-yellow color, and is confined to the inoculation stroke. (Eisenberg.)
Blood-serum.	Growth only occurs in the tract of the puncture, and forms a milk-white streak.
Temperature.	Grows at room and incubator temperature, but is very susceptible to high temperature. Destroyed in ten minutes at 60° C.
Rapidity of growth.	Grows slowly.
Formation of spores.	Sporulation (polar mode most probable) occurs at 32° to 40° C. Slower at 20° C., and below this none at all.
Aerobiosis.	Grows under mica plate.
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Sections are best stained with saturated alcoholic solution of methyl-blue, added to water until opacity occurs. Allow the sections to remain in the stain twenty to twenty-four hours, then wash in water (without acid), then in alcohol, and clear in cedar-oil. Are not colored by Gram's method. Cover-glass preparations are colored less intensely by the aniline dies than any similar organisms.
Pathogenesis.	Injected into the auricular vein of a rabbit, caused death in twenty-four to forty-eight hours (Fränkel, Simmonds). Guinea-pigs are killed by vaccination per os, as in cholera (Seitz). The bacilli were found in the internal organs, in the albuminous urine of very sick typhoid patients, in the blood (Neuhaus), and in the dejections of typhoid (Pfeiffer, E. Fränkel, Seitz).

112. Bacillus alvei. *Cheshire, Watson-Cheyne.*

Journal of the Royal Microscopical Society, Ser. II, Vol. V.

Place found.	"Foul brood,"—a disease attacking the larvæ of bees.
Form and arrangement.	Slender bacilli, with rounded or conical ends. In artificial cultures they often assume various sizes.
Motility.	Motile.
Growth.	
Gelatin.	*On Plates.*—Observed under a low power, a round or oval colony of bacilli is first seen, which, later, changes its shape to that of a pear, from the smaller extremity of which processes are thrown out. The bacilli grow along the stroke, with here and there small nodules, which send forth circuitous processes composed of bacilli, arranged in single file or in rows of several. These figures are very pretty. The gelatin is liquefied along the strokes, and canals are formed in which the bacilli move. *Tube Culture.*—At the entrance of the inoculation stab a delicate, ramifying growth occurs, and irregularly-shaped masses appear along the stab, which throw out threads with clubbed ends, giving a cloudy appearance to the gelatin. Evaporation occurs on the surface of the gelatin, accompanied by the formation of air-bubbles (as in Koch's comma bacillus). Rapid liquefaction begins, which has reduced the entire gelatin in two weeks. The liquid has a yellowish color and a peculiar odor.
Agar-agar.	*Tube Culture.*—At 37° C. the growth forms a whitish layer on the surfaces.
Potatoes.	At incubation temperature they form a dry, yellowish layer. Spores are formed.
Blood-serum.	At body temperature the growth is slow, and forms long spore-containing threads.
Temperature.	Best growth at 37° C. Does not grow under 16° C. In gelatin, best at 20° C.
Rapidity of growth.	Grows slowly. The rapidity of growth is in inverse ratio to the amount of material used.
Formation of spores.	Extraordinarily large spores, arranged in rows. The staves first swell, then become spindle-shaped. In opposition to most other spores, they are decolorized by tubercle-bacillus stain (Eisenberg).
Acrobiosis.	
Gas production.	
Gelatin reaction.	Very rapid liquefaction.
Aniline reaction.	Does not color with aniline colors; proportionately, the best stain is the methylene-blue and gentian-violet solution. They accept Gram's double stain.
Pathogenesis.	Half a syringeful of pure cultures injected into the subcutaneous tissue of a mouse's back caused its death in twenty-three hours. Œdema at the point of inoculation and in surrounding tissues. No change in the internal organs. Bacilli and spores are found in the œdema fluid, less in the heart-blood. Guinea-pigs inoculated die in six days with extensive necroses of the muscles and tissues, which contain caseous spots. The presence of the bacilli in these has not been positively proven.

113. Bacillus pneumonicus agilis. *Jens Schou.*

Fortschr. der Medicin. 1885. No. 15. Neumann, Zeitschr. f. klin. Med., Bd. XIII, S. 73.

Place found.	In the pneumonia which develops in rabbits after injury or section of the pneumogastric nerve (vagus-pneumonia).
Form and arrangement.	Elliptical cocci, diplococci, and short, thick bacilli.
Motility.	Motile.
Growth.	
Gelatin.	*On Plates.*—Round, dark, granular groups, with slightly roughened surface. Under a low power active motion is observed in the centre after twenty to twenty-four hours. The colonies are each surrounded by a circle of delicate processes, forming aréoli. *Tube Culture.*—In a few days the gelatin is liquefied, and the growth deposited as a thick, whitish sediment.
Agar-agar.	*Tube Culture.*—After twenty-four hours a somewhat prominent, moist, and transparent streak occurs along the inoculation stroke, from 1 to 2 mm. broad, which is not tenaciously adherent.
Potatoes.	Forms reddish, chamois colored colonies, which always remain flat, and which rapidly spread over the entire surface.
Blood-serum.	Grows very slowly and liquefies slightly.
Temperature.	Grows at room temperature.
Rapidity of growth.	Grows very rapidly.
Spore formation.	Not known.
Aerobiosis.	Facultative anaerobic.
Gas production.	Develops, on serum at 37° C., a disagreeable odor.
Gelatin reaction.	Liquefying very rapidly.
Aniline reaction.	Colors with aniline colors in the usual way, but not with Gram's method.
Pathogenesis.	Injected into the trachea, into the pleura and lungs, or inhaled, pure cultures cause rabbits to die with symptoms identical with those found in vagus-pneumonia. They were found in the blood. Besides the rabbit, the mouse is very susceptible; the guinea-pig in a less degree.

114. Bacillus of acne contagiosa of the horse. *Dieckerhoff, Grawitz.*

Virchow's Archiv, Bd. CII, S. 148.

Place found.	In the pus and scabs of the pustules occurring in "English pox," —acne contagiosa.
Form and arrangement.	Short staves, 0.2 m. long, straight, or slightly bowed.
Motility.	
Growth.	
Gelatin.	*Tube (Stab) Culture.*—Growth occurs sparingly along the puncture, at the entrance of which is a tuft the size of a millet-seed, of a pure-white color, or there may be many such bodies. *Tube (Stroke) Culture.*—On an oblique surface, round, knob-like bodies develop, which possess a peculiar arrangement of glistening spots upon their surface, occasioned by the superaddition of numerous small refractive bodies. This appearance is likened to that of shagreen.
Agar-agar.	*Tube Culture.*—As on blood-serum, but not so luxuriant.
Potatoes.	Growth is colorless and without odor.
Blood-serum.	After twenty-four hours at 37° C., whitish colonies arise along the inoculation stroke, about the size of poppy-seeds, which, later, assume a light-yellow or gray color. In the water of condensation a sand-like sediment settles to the bottom.
Temperature.	Best at 37° C.; under 17° C. vegetation ceases.
Rapidity of growth.	Grows rapidly on blood-serum; very slowly on other nourishing media.
Formation of spores.	
Acrobiosis.	The growth is very slight when access to air is cut off.
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	The best and quickest stain is aqueous solution of fuchsin. Aqueous solution of gentian violet is hardly so good, and Loeffler's strong alcoholic solution of methylene blue is almost without effect. An exceedingly pretty, blue-black coloring is obtained by Gram's method.
Pathogenesis.	Pure cultures rubbed into the intact skin of the horse, calf, sheep, and dog cause typical acne. In the rabbit, after subcutaneous inoculation, it causes erysipeloid swelling, hæmorrhages, general intoxication, necroses or burrowing pus-formations, according to the proportion of bacilli used. Applied to the skin of guinea-pigs, they caused death at the latest in forty-eight hours, with toxic symptoms. In mice the skin was refractory, but after subcutaneous inoculation symptoms of pyæmia and death supervened.

115. Micrococcus of mastitis of the cow. *Kitt.*

Zeitschr. f. Thiermed. u. Path., Bd. XII, S. 18.

Place found.	In the milky, pus-like products arising from inflammation of the milk-glands of the cow.
Form and arrangement.	Cocci, occurring singly, in pairs, or united in heaps, with here and there a tendency to the formation of chains. They have a diameter of 0.2 to 0.5 m
Motility.	
Growth.	
Gelatin.	*On Plates.*—Pearl-white, circular, sharply-defined, shining drops the size of a pin-head. *Tube Culture.*—Opaque, white, fungoid masses in the nail-culture form.
Potatoes.	Grayish-white, or muddy-yellow, quickly-growing colonies, which become lustrous after a few days, and take on the appearance of wax.
In milk.	Rapid growth when kept in the incubator. After six hours the milk has acid reaction.
Temperature.	Best growth at incubation temperature.
Rapidity of growth.	Grows very rapidly.
Formation of spores.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Stains well with all the aniline colors.
Pathogenesis.	Pathogenic to cows only, in which it causes a parenchymatous mastitis. Subcutaneous inoculation practiced on cows and on the smaller animals was without significance.

116. Micrococcus of lung-plague in cattle (pleuro-pneumonia contagiosa). *Poels, Nolen.* *Fortschr. der Medicin. 1886. S. 217.*

Place found.	In the lungs of cattle suffering from lung-plague.
Form and arrangement.	Cocci of varying size (0.9 m. in diameter), arranged singly and in twos and sixes, inclosed in distinct envelopes which are difficult to stain.
Motility.	
Growth.	
Gelatin.	*Plate Culture.*—Sharply-defined, round, white colonies of a lustrous, yellow color. *Tube Culture.*—Similar in appearance to the nail culture of Friedlaender's pneumo-bacillus; the head glistens slightly, and is of a cream-color.
Agar-agar.	*Tube Culture.*—Same as on gelatin, but growth is more rapid, especially at 37° C.
Potatoes.	Forms a moist, light-yellow coating.
Blood-serum.	At first the cream-color is not marked. This only occurs in the older cultures.
Temperature.	Best growth at 37° C.; resists temperature lower than 66° C., but if the atmosphere is laden with moisture it loses the power of development in fifteen minutes.
Rapidity of growth.	
Spore formation.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	Non-liquefying.
Aniline reaction.	Stained by all aniline colors, but with difficulty by Gram's method.
Pathogenesis.	Pure cultures, when injected into the lungs of rabbits, guinea-pigs, and dogs, cause pneumonic changes. Injected into the trachea, or by inhalation, they set up the same process in the dog. Injected into the lung of an ox, they cause an extensive pneumonic inflammation in seven days.

116*. Bacterium of hog-cholera (swine-plague—infectious pneumonia, Salmon). ***Billings, Dettmers.*** *Salmon, Second Annual Report of the Bureau of Animal Industry for 1885. Washington, 1886. S. Frank Billings, "Dr. E. Salmon's Swine-plague and Hog-cholera Critically Considered." Lincoln, Neb., 1889.*

Place found.	In the blood and organs of swine dying of hog-cholera.
Form and arrangement.	Ovoid cells, 0.8 to 1.2 m. long and 0.6 m. broad.
Motility.	Motile.
Growth.	
Gelatin.	*Plate Culture.*—Irregularly bounded colonies, the edges of which are sharp, their centres raised. In diaphanous light they have a bluish color; under low power they have the appearance of cloudy nets. *Tube Culture.*—Along the stab, white, irregular, round, crystalloid colonies. Upon the surface, a white, milky, rapidly-spreading coating, the periphery of which is often serrated.
Agar agar.	*Tube Culture.*—A white coating along the stroke.
Potatoes.	A coffee-brown layer, resembling the growth of bacillus malle, with sharply-defined edges.
Temperature.	Grows at room temperature.
Rapidity of growth.	Grows rather rapidly.
Spore formation.	Not known. Multiplies by segmentation. Bears drying for a long time.
Aerobiosis.	Does not grow under anaerobic conditions.
Gas production.	Not observed.
Gelatin reaction.	Does not liquefy.
Aniline reaction.	Are stained by the common aniline colors, but usually the ends are more intensely stained than the middle. Are not stained by Gram's or Weigert's method.
Pathogenesis.	Mice, rabbits, and guinea-pigs are very susceptible to subcutaneous inoculation; pigeons are less susceptible, while chickens, sheep, and calves are refractory. The animals die in from eight to sixteen days; there frequently occurs a slight local reaction; the spleen, liver, and kidneys are enlarged, while hæmorrhages occur in all the internal organs. Swine react best after being fed upon cultures of the bacilli following a forced fast of several days. Under these conditions large areas of a necrotic process are seen in the mucous membrane of the large intestine, and inflammation and occasionally ulceration of the stomach and ileum.

116**. Bacteria of Texas fever. *Billings.*

The Southern Cattle Plague (Texas fever). Lincoln, Neb., 1888.

Place found.	In the blood, gall, urine, and organs of animals sick or dying of Texas fever.
Form and arrangement.	Ovoid cells, about twice as long as they are broad. Their length is about one-sixth the diameter of a red blood-corpuscle. Besides these typical forms, there are others which vary in size and form.
Motility.	Possesses rotary and scintillating movements.
Growth.	With the exception of very slight variations, the same as the bacteria of swine-plague.
Gelatin.	
Agar-agar.	
Potatoes.	
Blood-serum.	
Temperature.	The same as bacteria of swine-plague.
Rapidity of growth.	
Spore formation.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	
Aniline reaction.	Nuclear stains are taken up quicker by and color deeper the ends of the bacteria, while the middle remains colorless.
Pathogenesis.	Small animals, inoculated by pure cultures, suffer from fatal septicæmia. An inoculated steer died after an illness lasting five months and resembling Texas fever. The section showed catarrhal inflammation of the stomach and intestines; parenchymatous changes in the heart, liver, kidneys; the spleen was much enlarged.

II.

PATHOGENIC BACTERIA.

B. Not Cultivated Outside the Animal Body.

117. Spirochæte Obermaieri.

Cent. f. d. med. Wissensch. 1873. XI, 10.

Place found.	In the blood of patients afflicted with recurrent fever.
Form and arrangement.	Spiral threads, 16 to 40 m. long, with "screw threads" of equal arrangement. The flagellum is distinctly visible. (Koch.)
Motility.	Very motile by means of wavy undulations.
Growth.	Has not as yet been cultivated successfully outside the animal body, although in blood-serum they preserve their movements for some time.
Temperature.	
Rapidity of growth.	
Spore formation.	
Aerobiosis.	
Gas production.	
Gelatin reaction.	
Aniline reaction.	After "fixing" in the flame, cleanse with 4 to 5 per cent. acetic-acid solution and stain with the ordinary aniline solution.
Pathogenesis.	Found in the blood of those suffering from recurrent fever. Apes inoculated with this blood are found to contain the organisms circulating in the blood. The animals were killed at the climax of the disease. Carter: *D. Med. Wochenschr.*, 1879, Nos. 16 and 25; Koch: *Mitth. a. d. Kais. Ges.-Amt.*, Bd. 1, S. 40.

118. Syphilis bacillus. *Lustgarten.*

Med. Jahrb. d. k. k. Ges. d. Aerzte. Wien, 1885.

Place found.	In the tissues and secretions arising in syphilis.
Form and arrangement.	In size and shape they resemble the tubercle bacillus, frequently bent or curved, or slightly S-shaped. Their ends are often thicker or clubbed, and their outlines seem broken.
Motility.	
Growth.	Not cultivated as yet outside the animal body.
Temperature.	
Rapidity of growth.	
Spore formation.	The bacilli contain two to four bright, shining, oval spots, probably spores.
Aerobiosis.	
Gas production.	
Gelatin reaction.	
Aniline reaction.	Lustgarten: Sections are placed for twelve to twenty-four hours in the following solution at room temperature, after which it is warmed for two hours at 40° C.: solution of gentian violet 11, aniline water 100. The sections are then placed for a few minutes in absolute alcohol, and from there transferred to a 1.5 solution of permanganate of potassium. After ten minutes they are immersed in a solution of c. p. sulphuric acid for a moment, then rinsed in distilled water. If sections are not discolorized this process must be repeated. Dehydrate with alcohol and mount in Canada balsam. De Giacomi: Twenty-four hours in fuchsin solution, rinse in water, then in solution of chloride of iron, alcohol, clove-oil.
Pathogenesis.	The bacilli do not occur free in the tissues, but inclosed in cells, which are round, oval, or irregularly polygonal, enlarged to twice the size of a white blood-corpuscle. Occur most numerous when only a short time has elapsed since the infection, and especially when the primary infiltration is young.

118*. Plasmodium malariæ. *Laveran, Marchiafava, Celli, Guarnieri, Osler.* *For full bibliography, see Archives de Méd. Experimentale.*

Place found.	In blood-corpuscles of those suffering from malarial poisoning.
Form and arrangement.	Protoplasmic corpuscles of varied form (round, oval, crescentic bodies and flagella), with or without pigment, as the malarial process in which they occur is chronic or acute, due to changes occurring in the hæmoglobin. They present a cortical substance (ectoplasma), surrounding a less colored portion (endoplasma), which contains sometimes one or more nuclei and nucleoli.
Motility.	Rapid amœboid movements.
Growth.	A method by which these organisms may be cultivated outside the human body has not as yet been found. They probably need a vital nutritive media.
Spore formation.	It multiplies by cleavage of a whole or a part of the protoplasmic substance. It becomes stationary, and the pigment of those so supplied accumulates in the centre, while new elements bud from the periphery, which elements may from the first be pigmented, and may possess undulating flagella.
Aniline reaction.	The organisms may be observed when the blood is fresh. A strong lens is required (Zeiss, $\frac{1}{12}$ to $\frac{1}{18}$ homogeneons immersion) to detect them. Dry preparations may be beautifully stained by methylene blue.
Pathogenesis.	The blood of a person suffering from malaria inoculated into the blood of a second individual produces a typical malarial attack, and the organisms multiply and are capable of producing progressive infection. They disappear rapidly under the administration of quinine. Celli and Guarneri believe the organism belongs to the genus Sporozoa (protozoa), family Gregarinidæ, and order Coccidium.

III.

FUNGI.

119. Actinomycoses.

Bollinger, Centr.-Bl. f. d. med. Wissenschaft. 1877. No. 27. Israel, Virchow's Archiv, Bd. LXXVII u. LXXVIII.

Place found.	In actinomycosis occurring in man and cattle.
Color of growth.	
Mycel arrangement.	The parasite is a round, seldom oblong, sometimes flattened, hollow body, with a small central cavity and a relatively thick wall, made up of pyriform or club-shaped elements arranged perpendicular to the surface. The arrangement gives it the form of a rosette. The club-shaped elements are separated with difficulty. The parasites are sometimes calcified.
Fructification organs.	
Growth.	Typical cultures were obtained after five to six days on cattle serum, agar-agar, and gelatin, without liquefaction. The centre of the culture consists of small, yellowish-red, nearly round spots, which are provided at their periphery with extremely delicate, cloudy-looking, ramifying processes. By degrees the small, yellowish-red masses increase in number, and are provided with a flaky-white covering. (Boström.) Easily procured when conditions for anaerobic existence is provided according to Buchner's method.
Agar-agar.	In forty-eight hours at 36° C. a slight prominence at the point of inoculation occurs; this increases in size until in several weeks it presents the appearance of a rather large, yellowish-white body, which grows into the agar and is with difficulty separated from it. (Bujwid.)
Temperature.	Grows only at cultivation temperature.
Examination methods.	Very difficult to stain. Sections are best stained by Gram's or by Weigert's method: first in orseille, then in 1-per-cent. aqueous gentian-violet solution, whereby the central thread-work is colored blue and the periphery a ruby red.
Pathogenesis.	Inoculation subcutaneously in calves was frequently successful. Feeding was without effect. Dogs are not susceptible. Lately, Israel has injected pure cultures into the abdominal cavity of rabbits with typical pathological results. Test experiments have been frequently made by Bostrom.

120. Penicillium glaucum.

Place found.	Spores are present everywhere. It is the most common mold.
Color of growth.	Green.
Mycel arrangement.	Arranged horizontally, are straight or slightly undulating and continuous. The fruit hyphæ are arranged perpendicularly.
Fructification organs.	The fruit hyphæ are divided at their terminal ends in a forked form (basidien), from which spring fine processes (sterigmata) which give off the greenish conidia.
Growth.	Whenever mold formations may exist.
Temperature.	Optimum for mycelium formation, 26° C.; that for conidia, 22° C. (Wiesner). Room temperature is most suitable; no growth at all at body temperature.
Examination methods.	Since mold fungi are not moist, it is well to first treat with alcohol to which some (3 to 4 drops) ammonia has been added, and they are allowed to sink to the bottom. They may then be preserved in glycerin or stained with aniline color; the best is Loefler's methylene blue, whereby the mycel and fruit-bearers are brought into prominence, while the spores remain unstained.
Pathogenesis.	Non-pathogenic.

121. Aspergillus glaucus. *de Bary.*

Place found.	On sugar fruits.
Color of growth.	Green.
Mycel arrangement.	As in No. 120.
Fructification organs.	Fruit hyphæ are much longer than those of No. 120, but do not ramify at the end. A mace-like enlargement occurs here, instead, from which ray-like, short, club-shaped sterigmatæ arise, which give off the spores from their long extremity.
Growth.	It shows a special preference for the juice of fruits.
Temperature.	Thrives well at 10° to 15° C. Disappears at 25° C. (Siebenmann.)
Examination methods.	
Pathogenesis.	Not pathogenic. Has often been described as a pathogenic, but was then confounded with aspergillus fumigatus.

122. Aspergillus niger.

Place found.	
Color of growth.	Black.
Mycel arrangement.	The same as No. 121.
Fructification organs.	
Growth.	
Temperature.	Optimum of mycel as well as conidia formation, 34° C. (Raulin.)
Examination methods.	
Pathogenesis.	

123. Aspergillus fumigatus.

Lichtheim, Berl. klin. Wochenschr. 1882. No. 9 u. 10. Gaffky, Mitth. a. d. Kais. Ges.-Amt., Bd. I, S. 126. Schütz, ebendaselbst, Bd. II, S. 208.

Place found.	In the air-passages of a bird (Fresenius). On white bread (Lichtheim). Observed saprophytically in human lungs, external auditory meatus, etc. (Pierce).
Color of growth.	At first blue-green, very like the color of penicilium glaucum. Older cultures assume a dirty, grayish color.
Mycel arrangement.	Smaller in all parts than No. 121.
Fructification organs.	Spores only one-quarter as large as the conidia of No 121.
Growth.	Best growth occurs on bread.
Gelatin.	Under low power the spore-bearing sterigmen are seen actively engaged in fructification, giving the appearance of a thorn-apple.
Temperature.	Optimum for fructification, 37° to 40° C. (Lichtheim.) Minimum, 15° C. (Fresenius.)
Examination methods.	
Pathogenesis.	Dogs and rabbits, into whose blood-vessels the spores have been injected, present peculiar disturbances of co-ordination, the cause of which is localization of the fungi in the membranous labyrinth. The animals die in twenty-four hours. The fungi are found germinating in the various organs, especially in the kidneys and heart-muscle. Small pieces of the organs placed in sterilized bread cause a characteristic growth of the aspergillus. It is not to be presumed that the human organism is immune.

124. Aspergillus flavescens.

Place found.	On white bread.
Color of growth.	Yellowish green.
Mycel arrangement.	Smaller than No. 121, but larger than No. 123.
Fructification organs.	Spores one-half the size of No. 121.
Growth.	Grow best on bread.
Temperature.	Best growth at breeding temperature.
Examination methods.	
Pathogenesis.	Like No. 123.

125. Aspergillus subfuscus. *Olden, Gade.*

Norm. med. Arkiv. 1886. Bd. XVIII, No. 9.

Place found.	On white bread.
Color of growth.	Olive-yellow, bordering on black.
Mycel arrangement.	Mycels are snow-white, evenly spread on the substrata, with rather thick threads, having the same diameter as the fruit hyphæ.
Fructification organs.	Fruit hyphæ 300 to 400 m. long and 10 to 20 m. broad, with globular heads about 30 m. in diameter. From these the sterigmata radiate. Their pointed ends give off small greenish-black spores.
Growth.	Best growth occurs on acidulated nourishing media.
Temperature.	Optimum at 37° to 38° C.; thrives also at 15° to 20° C., but slowly.
Examination methods.	
Pathogenesis.	When introduced into the circulation of rabbits and cats, they cause a general mycosis, from which the animal dies. The process is less severe than those caused by Nos. 123 and 124. The stomach and alimentary canal are sometimes attacked by the fungus.

126. Aspergillus nidulans. *Eidam, Lindt.*

Archiv f. exp. Path. u. Pharmak., Bd. XXI, S. 284. Beitr. z. Biol. d. Pflanzen, Bd. III, S. 392.

Place found.	On white bread.
Color of growth.	Forms a pretty, chlorine-green, delicate layer, 1 mm. in depth.
Mycel arrangement.	Mycels possess hyphæ with many ramifications. In older cultures thick, white mycels, which frequently have a rosy-red color, giving them a soiled appearance.
Fructification organs.	Fruit-bearers are at first colorless and are not ramifying. Later they color brown and become branched. Ramifying sterigmata rest on hyphæ that are at first club-formed and later more triangular. The chains of conidia in their arrangement resemble at first the head of a Medusa, and, later, collect and form a long cylinder. Spores are round, very small (3 to 4 m.). In old cultures perithecien and ascospores are formed.
Growth.	Grows on bread-infusion, agar-agar, potatoes, bread. Has a peculiar circular or concentric form. The coloring matter that is formed colors the potato a dark, brownish red. This discoloration is visible also on bread. Agar-agar remains uncolored.
Temperature.	Grows best at 40° C.
Examination methods.	As in No. 123.
Pathogenesis.	Death results in a rabbit sixty hours after the spores are injected into the ear-vein. On section the kidneys are found to be enlarged to twice their original size, and are filled with fungous foci. Numerous foci are also found in the heart-muscle. The fungus produces yellow conidiæ in the animal body. They are less pathogenic than Nos. 123 and 124, inasmuch as only a relatively small number reached fructification.

127. Mucor mucedo.

Place found.	Generally on animal excrements, especially horse manure.
Color of growth.	White.
Mycel arrangement.	Form of widely ramifying threads consisting of single cells.
Fructification organs.	Undivided fruit-bearers ending in an enlargement (antheridium), which, when ripe, forms darkly-colored parenchyma. Large, oval, brown-colored spores are formed in its interior, which escape by bursting their envelope.
Growth.	Best growth on acid media. The pure culture gives a layer an inch in thickness, which is composed of threads arranged at right angles with each other, the ends of which are surmounted by a round body the size of a poppy-seed.
Temperature.	Grows at body temperature.
Examination methods.	
Pathogenesis.	Non-pathogenic.

128. Mucor rhizopodoformis. *Lichtheim.*

Zeitschr. f. klin. Med., Bd. VII, Heft 2.

Place found.	On white bread.
Color of growth.	Snow-white; later, mouse-gray.
Mycel arrangement.	Forms colorless, disconnected mycels, growing into the substrata.
Fructification organs.	Fruit-bearers have the appearance of tufts, which are attached to the nourishing media by special hair-like roots. Sporangia regular, globular expansion; spores globular.
Growth.	Grows best on bread-gelatin infusion, which it liquefies. It forms a solid, blackish-gray, fungoid scum.
Temperature.	Best at 37° C.; in room temperature; also development between 12° to 15° C.
Examination methods.	In fresh sections the fungus is easily observed after treating with acetic acid or solution of potassium. Pretty preparations are made by staining with picrocarmine and preserving in glycerin. The mycelium cannot be colored separately, except by Ehrlich's hæmatoxylin solution. They then assume a blue color.
Pathogenesis.	Spores have been injected into the circulation of rabbits. For twenty-four hours after the injection a period of latency is observed, then death follows in forty-eight to seventy-two hours. On section we find evidences of a high grade of disease. The kidneys are enlarged to double their normal size, and within them herds of fungi are found larger than those occurring after inoculation with aspergillus. There are striking changes in the lymphatics of the alimentary canal. It is often possible to demonstrate the presence of the fungi in the spleen and bone-marrow, exceptionally in the liver. They are numerous in the diseased portions of the intestines. All other organs remain unaffected. This is especially true of the striped muscles, differing in this from the aspergillus, which seems to have a selective affinity for the striped muscle. When animals retain life for a greater period (eight to fourteen days) the fungus is sometimes found in the lungs. Dogs are immune. It is doubtful whether the fungi can grow in the human body.

129. Mucor corymbifer. *Lichtheim.*

Place found.	Is an infrequent contamination of gelatin-bread infusion; otherwise, of seldom occurrence.
Color of growth.	The gray color is not so deep as that of No. 128, on account of the small, colorless sporangium.
Mycel arrangement.	Loose, curly mycel, with smaller germinal sacs than No. 128.
Fructification organs.	Fruit hyphæ are umbellate, resembling a bunch of grapes, and have pear-shaped sporangia. The spores are oblong and smaller than those of No. 128.
Growth.	On gelatin-bread infusion forms a moldy scum, the texture of which is looser than No. 128.
Temperature.	Best growth at 37° C. In room temperature the growth is less rapid than No. 128.
Examination methods.	In contrast with No. 128 the germinal sac stains with methylene blue and like coloring agents.
Pathogenesis.	Less malignant than No. 128.

130. Mucor pusillus. *Lindt.*

Archiv f. exp. Path. u. Pharm., Bd. XXI, S. 272.

Place found.	On moist, white bread.
Color of growth.	Snow-white; later, mouse-gray.
Mycel arrangement.	Mycel continuous, without septi, very fine and delicate. They ramify in every direction, and form a thin, velvety layer. The mycelium send forth numerous short hyphæ on the nourishing substrata, and numerous additional continuations up into the air; but true air-mycels are wanting. All parts of the fungi are distinguished by their delicacy.
Fructification organs.	Possess numerous sporangia-bearers, 1 mm. in height, with globular, black sporangia, surrounded by a prickly sclerotium. They possess, toward the fruit hyphæ, a sharply-defined, oval, or globular columella. The spores are very small (3 to 3½ m.), globular, colorless, and are surrounded by a delicate membrane.
Growth.	Best on agar-agar-bread infusion. At first is sharply defined at the point of inoculation, and, later, spreads over the whole plate.
Temperature.	Thrives only at a high temperature, the lowest being 24° to 25° C.; maximum, above 50° C.; optimum, 45° C.
Examination methods.	It cannot be stained trustworthily by aniline colors, nor with Lichtheim's acid hæmatoxylin solution. The presence of the fungi is best ascertained in sections which have been placed in Müller's fluid.
Pathogenesis.	Spores injected into the blood-vessels of rabbits caused death in two and a half to four and a half days. The pathogenic and anatomical picture is the same as that caused by Nos. 128 and 129, but is of a more benign character. The pathogenic process occasioned by the mucores is the same in every instance, and the same organs are affected in the same typical order: kidneys, alimentary canal, mesentery glands, and spleen.

131. Mucor ramosus. *Lindt.*

Archiv f. exp. Path. u. Pharm., Bd. XXI, S. 275.

Place found.	On moist, white bread.
Color of growth.	Snow-white; later, gray-brown; somewhat darker than No. 129.
Mycel arrangement.	Mycelium very delicate, sending forth atmospheric hyphæ 3 to 6 mm. in height.
Fructification organs.	Fruit hyphæ similar to No. 129, dividing dichotomously, with pear-formed sporangia; columella similar to No. 129; sometimes spherical at the vertex. Spores colorless, with delicate, smooth walls, markedly oval, and measuring 3 to 4 m. in width and 5 to 6 m. in length.
Growth.	In agar-agar-bread infusion, mostly confined to the inoculation stroke at first; later, spreading over the whole plate. Growth is excellent also on potatoes.
Temperature.	Best at 40° C.; also in room temperature at 15° to 16° C., producing in five or six days a fructifying mycel.
Examination methods.	As in No. 130.
Pathogenesis.	Pathogenesis similar to that produced by the other mucores, but the fatal issue is more rapid on account of the rapidity with which it develops in the organism. The mesenteric glands, Peyer's patches, and kidneys are especially diseased, and have a marked hæmorrhagic character.

132. Achorion Schönleinii (favus fungus).

Grawitz, Virchow's Archiv, Bd. CIII, S. 402.

Place found.	In the scales and scabs from those suffering from favus.
Color of growth.	White, or yellowish.
Mycel arrangement.	Rather extensive ramifications; flat, distinct hyphæ; distinguished by the ramifications occurring at acute right angles.
Fructification organs.	Special fruit hyphæ are wanting; yet, under certain conditions (serum at 30° C.), small, ellipsoid conidia are observed, 5.2 m. to 6.5 m. in diameter.
Growth.	
Gelatin.	*Plate Culture.*—Growth slow, in form of white, round masses, situated in foci of liquefied gelatin. Mycel sterile. *Tube Culture.*—Very poor, conidia not formed.
Agar-agar.	At 30° C. similar to No. 129.
Blood-serum.	30° C. the best nourishing media.
Milk.	No growth occurs. Fungi destroyed.
Temperature.	Completely destroyed at room temperature.
Examination methods.	
Pathogenesis.	At the stage of conidia genesis it produces true favus.

133. Trichophyton tonsurans (fungus of herpes tonsurans). *Eichstädt.*

Grawitz, Virchow's Archiv, Bd. CIII, S. 402.

Place found.	In the scales and scabs from persons afflicted with herpes tonsurans.
Color of growth.	Yellowish white.
Mycel arrangement.	Divided into many ramifications, flat, with distinctly articulate hyphæ.
Fructification organs.	Possesses no special organs of fructification; yet, under given conditions (serum 30° C.) we may observe the breaking up of the mycel into small, rounded conidia 6.5 m. in diameter.
Growth.	Slower growth than No. 132; more rapid than No. 128.
Gelatin (7½ per cent. alkaline).	*Plate Culture.*—At 30° C., white, circumscribed foci develop, having a somewhat acutely-prominent centre. The mycel-threads radiate over the gelatin. Older cultures assume an orange color.
Gelatin.	*Tube Culture.*—Gelatin rapidly liquefies. The growth floats on the surface. It is thick and arranged in two layers, the upper one white, the lower yellow. The clumps which cling to the tube-wall have dark-yellow centres and radiating threads form their periphery.
Agar-agar.	*Tube Culture* (at 30° C.).—At first the growth is similar to No. 132. Later, distinct differences develop; lentel-shaped spots arise in the clumps of mycelium which become orange-yellow, while a flour-like dust arises on their surface.
Blood-serum.	*Tube Culture* (at 30° C.).—Invades the entire surface. When the tube is held to the light the coating seems to be made up of coalesced round clumps. After a few days the growth assumes a yellow color and the serum is liquefied. The development on serum is not, however, complete or rapid.
Milk.	Is destroyed.
Temperature.	Growth is retarded in cool room temperature. Best growth at 30° C.
Examination methods.	
Pathogenesis.	At the stage of conidia generation it causes, in man, herpes tonsurans.

134. Fungus of tinea galli (chicken-itch). *Gerlach.*

Schütz, Mitth. a. d. Kais. Ges.-Amt., Bd. II, S. 224.

Place found.	From scabs removed from the combs of chickens affected with the so-called "white-comb."
Color of growth.	White.
Mycel arrangement.	Consists of articular threads, often ramifying, and of variable sizes.
Fructification organs.	Fruit-bearers are wanting. Conidia are, perhaps, present.
Growth.	Best growth occurs on bread-infusion at 30° C.; it occurs as pale-white, glistening layers or plaques, and produces dark-red coloring matter which diffuses through the nourishing media. It also occurs on gelatin, which it liquefies; also very well on agar-agar and potato.
Temperature.	Best growth at 30° C.
Examination methods.	
Pathogenesis.	Inoculations of pure cultures on chickens produce the characteristic disease. Mice, rabbits, and other test animals are immune.

135. Fungus of mouse-favus. *Nicolaier.*

Flügge, Mikroorganismen, S. 100.

Place found.	Accidentally.
Color of growth.	Pure white; later, reddish to reddish brown.
Mycel arrangement.	Dense, thin mycel with delicate hyphæ superimposed.
Fructification organs.	Special organs of fructification or distinct spore formation not as yet observed.
Growth.	Development occurs on acidulated agar-agar and on potatoes acidulated with acetic acid, having the appearance of a crust of sugar.
Temperature.	Best growth at 30° to 35° C.
Examination methods.	
Pathogenesis.	Inoculated with small masses of pure culture, mice become affected with a peculiar disease. A scab forms on the head of the animal, which enlarges and spreads until it becomes an immense grayish, whitish mass of scales. Inoculation on a rooster was without effect.

136. Oidium lactis.

Grawitz, Virchow's Archiv, Bd. CIII, S. 402.

Place found.	In sour milk and butter.
Color of growth.	White.
Mycel arrangement.	Closely arranged mycelium from which spring undulating and bent threads.
Fructification organs.	No antheridium, but rows of cylindrical conidia spring directly from the filaments arising from the mycelium. It forms the boundary-line to the unicellular fungi, which propagate by budding (yeast-fungi).
Growth.	
Gelatin.	*Plate Culture.*—Grows at 20° C. on alkaline 7½-per-cent. gelatin. Its growth is the most rapid and luxuriant of the fungi, with the exception of Nos. 132 and 133. It covers the whole surface of the plate with delicate white, long, hair-like mycelium, and emits an odor resembling that of sour milk. *Tube Culture.*—Sends forth long, delicate threads through the gelatin, but does not liquefy it.
Agar-agar.	*Tube Culture.*—At 30° C., forms fine, white, stellate bodies, which coalesce in an even, delicate film, which spreads over the surface. At the latter point it assumes a shiny character.
Blood-serum.	*Tube Culture.*—At 30° C., diffuse vegetation spreads evenly over the surface. No limited foci.
Milk.	Forms a moldy skin.
Temperature.	Best growth between 15° to 20° C. Growing faster than Nos. 133 and 134.
Examination methods.	
Pathogenesis.	Non-pathogenic. They have been inoculated by subcutaneous inoculation and injections into the subcutis, and directly into the blood-passages, but without effect (Hüppe).

137. Oidium albicans (Soor).

Grawitz, Virchow's Archiv, Bd. LXX. Baginsky, Deutsche Med. Wochenschr. 1885. S. 866.

Place found.	From the mucous membrane of the mouths of infants.
Color of growth.	
Mycel arrangement.	Consists of oval or cylindrical cells, 3 to 5 m. in length to 5 m. in thickness. Develops like the saccharomycetes, or in long, thread-like mycelii.
Fructification organs.	
Growth.	
	Plate Culture.—Not liquefying. Forms snow-white colonies.
Gelatin.	*Tube Culture.*—White or light-yellow groups occur through the gelatin. Those toward the bottom have delicate, ray-like continuations; the same toward the surface are club-shaped at their ends. The deepest situated develop thread-like mycelium.
Potato.	Growth is rapid, and forms a thick, white, yeast-like covering, made up of little heaps, varying in size from a hemp-seed to that of a lentil. The spores occur in chains.
Bread.	Growth from the inoculation stroke over the surface is veil-like and nearly snow-white.
Temperature.	
Examination methods.	
Pathogenesis.	According to Klemperer, pathogenic to rabbits, which die within twenty-four to forty-eight hours after intra-venous injections of pure culture. The organs are crowded with long threads of mycelium.

138. Saccharomyces rosaceus (yeast-fungi).

Place found.	Air.
Color of growth.	Pink.
Mycel arrangement.	No mycelium.
Fructification organs.	Proliferate from spore chains (yeast-chains).
Growth.	
Gelatin.	Non-liquefying. Pink colonies grow on surface.
Agar-agar.	Same as on gelatin.
Temperature.	Best growth at room temperature.
Examination methods.	Is stained the same as the bacteries, with aniline colors.
Pathogenesis.	Not pathogenic.

APPENDIX.

BACTERIOLOGICAL TECHNIQUE USED IN THE CULTIVATION AND STAINING OF BACTERIA.

NOURISHING MEDIA.

Fluid Nourishing Media.

Meat-water Bouillon, after Loeffler (1).

One-half kilo of finely-chopped lean meat is added to a litre of water; this mixture is usually allowed to stand for twenty-four hours in a cold place, or it may be placed in a water-bath for three fourths of an hour and then filtered. Add 10 grammes dry, powdered peptone and 5 grammes table salt. Warm to cooking-point, and add gradually saturated solution of sodium carbonate until red lacmus-paper shows a light-red color and the blue turns slightly red. Cook for one or two hours, until the separable albuminous matter is precipitated from the clear fluid. After cooling, the whole is filtered through a dampened filter. An almost colorless, weakly, alkaline bouillon is the result, which must not become turbid after sterilization.[1] If it does become cloudy it must be again filtered.

Meat-extract Bouillon, after Hueppe (2).

One litre of water, 30 grammes powdered peptone, 5 grammes grape- or cane-sugar, and 5 grammes meat-extract (instead of peptone and meat-extract 20 to 30 grammes meat-peptone may be used). Cook, neutralize, and proceed as in the preceding preparation, except that this must be exposed to the sterilization process for a longer time, as the meat-extract contains innumerable spores. The resulting bouillon is always of a yellow color.

Milk.

Fresh milk, contained either in cotton-corked vessels or in double-saucers, is exposed to 120° C., in steam under pressure, for ten to fifteen minutes; or, in a current of steam at 100° C. for twenty to thirty minutes, for three successive days.

Solid Media.

Ten-per-cent. Meat-water Peptone Gelatin, after Koch and Loeffler (1).

To 1 litre of meat-water bouillon (either cooked or allowed to stand twenty-four hours), 100 grammes gelatin, 10 grammes peptone, 5 grammes table-salt are added, the whole shaken and warmed on water-bath until the gelatin is melted. Neutralize as in No. 1: cook for one-half hour in Koch's steam-sterilizer until the fluid is clear and the separable albumen coagulated; filter hot through a food-filter or hot-water filter. The gelatin so produced must have a weakly alkaline reaction, must be clear as water, of a very high amber color, and the "control tubes" must remain sterile. If a slight cloudiness does occur from any cause other than a wrong reaction or insufficient cooking, it is easily cleared by adding the white of one or two eggs, or 10 to 20 cubic centimetres blood-serum, cooking for one-fourth of an hour, and filtering as before.

[1] Sterilization of all nourishing media, where not otherwise specified, is accomplished by cooking for fifteen or twenty minutes on three successive days in Koch's steam-sterilizer. The first or even second cooking does not always kill the spores; hence the necessity for three cookings.

Ten-per-cent. Meat-extract Peptone Gelatin, after Hueppe (2).

One litre of water, 100 grammes gelatin, 30 grammes peptone, 5 grammes grape-sugar, and 5 grammes meat-extract, treated as the preceding. The resulting gelatin is brownish yellow. Sterilize eight times on account of meat-extract.

Wort Gelatin.

Especially good for acid media used in the culture of fungi. Beer-wort, mixed with 10-per-cent. gelatin, liquefied by warming, and the mixture is cooked several times in steam-sterilzer, after which it is filtered without neutralizing.

Milk-serum Gelatin, after Raskina (3).

(*a*) Substitution of the Casein by Peptone.—One litre of unskimmed milk is warmed to 60° to 70° C., and then 70 to 100 grammes gelatin added. After the gelatin has melted the whole is cooked several minutes, whereby the casein is precipitated. The cloudy fluid is heated about twenty minutes at incubation temperature to allow the fat to accumulate upon the surface. It is then allowed to cool and afterward skimmed. The nearly clear fluid is now cooked, with 1-per-cent. peptone added, neutralized, and then filtered.

(*b*) Substitution of the Casein by Sodium-egg-Albumen.—To the whites of eggs is added drop by drop a concentrated solution of sodium, until the albumen is a nearly solid, transparent mass. This is cut up with a sterilized knife, washed with sterilized distilled water, and then allowed to stand until it is changed to a thick, yellowish fluid. Three per cent. of this strongly-alkaline fluid is added to milk-serum gelatin (*a*) instead of the peptone. Any particular neutralization is unnecessary.

(*c*) Milk-Casein Gelatin (containing casein).—Milk is left for two days, then skimmed and the coagulum warmed twenty minutes at 70° C., whereby the serum is more easily pressed out. The well-squeezed casein coagula is then washed in 95-per-cent. alcohol, dried, pulverized, and the fat extracted by exposure to ether in a bulb. The casein is then dried between filter-paper and heated for ten to fifteen minutes at 120° to 140° C. The casein is then found transformed to a sticky, tenacious mass, which, after washing with natrium solution becomes horny, transparent, and, after drying, becomes hard as stone. Milk-serum gelatin is mixed with 2.5 per cent. of this preparation of casein.

Lactic-acid Gelatin, after Marpmann (4).

One litre of milk is heated to boiling and then added to the same amount of a weak solution of sulphuric acid until precipitation occurs. The resultant transparent serum is freed from the flocculent albuminous bodies by straining through a woolen collatorium, mixed with pure calcium carbonate to oversaturation, and then cooked once. The clear fluid, which must have a pronounced alkaline reaction, is decanted off and 10-per-cent. gelatin is dissolved therein. Sterilize by heating in dry-oven for one hour at 80° C. for eight days.

Potato Gelatin, after Holz (5).

Potatoes are pared and thoroughly cleansed and then grated on a common kitchen-grater. The juice from this pulp is pressed through a clean woolen cloth, and allowed to stand for twenty-four hours at 10° C. in a bulb, the mouth of which is closed by a cotton plug. A smutty-brown fluid is the result. After twenty-four hours it is filtered and the filtrate is heated in steam-sterilizer for one-half hour and then again filtered; 400 grammes of the resulting clear potato-juice are mixed with 40 grammes of gelatin and heated in the steam-sterilizer for three-fourths of an hour, filtered, and then poured into whatever vessel desired. Sterilize by heating in steam-sterilizer for one-fourth of an hour on three successive days. The potato gelatin so procured is clear, transparent, and of a brownish color.

Two-per-cent. Meat-water Peptone Agar-agar, after Koch.

To 1 litre of meat-broth (procured either by heating or allowing it to remain in a cool place) add 10 grammes peptone and 5 grammes of salt, and filter. It is then cooked for one

hour, filtered, and to the filtrate is added 20 grammes finely-cut agar agar, which has before been soaked for twenty-four hours in water and squeezed through a cloth just before using. The whole is then neutralized by adding several drops of a concentrated solution of natrium carbonate. It is then heated over a common water-bath, in which case care must be taken that the water of condensation is replaced; or it may be placed in the steam-sterilizer for an hour, until the albuminous bodies are precipitated in a flocculent mass from the clear agar solution. This precipitation is accelerated by adding the whites of one or two eggs. The filtration can be accomplished only by the aid of the hot-water filter, and, if the mixture has been insufficiently cooked, or the separation of the albumen has not thoroughly taken place, the filtration is rendered still more tedious, even if this takes place in the steam-sterilizer. Sterilize as with bouillon. The finished agar-agar is, in the fluid state, clear and white as water; when hardened, it is somewhat opaque.

After Richter (6).

The agar-agar is soaked for two hours over a water-bath in about 300 cubic centimetres Mosel wine; the agar is quite soluble in this warm wine. This solution is at once sterilized and added to meat-broth containing peptone and salt, and filtered in the hot water filter.

Two-per-cent. Meat extract Peptone Agar-agar, after Nocard and Roux (7).

Simply add 6- to 8-per-cent. glycerin to the agar-agar produced by the methods already mentioned.

Media according to Kowalski (8).

One kilo of calve's lung is chopped very fine, then placed in 2 litres of distilled water, cooked, and after one half hour filtered and pressed in a clean piece of linen. To the resultant fluid add chloride of sodium, 18 grammes; sulphate of natrium, 25 grammes; phosphate of kalium, 9 grammes; sugar, 90 grammes; sulphate of ammonia, 9 grammes; peptone, 25 grammes. After complete solution has taken place, 10- to 15-per-cent. gelatin or 2-per-cent. agar is added, the latter ingredient having been soaked in water for twenty-four hours. The mixture is then dissolved by cooking and an occasional active shaking. It is then neutralized by a solution of equal parts of potassium and sodium carbonates, brought up to 2½ litres by addition of water, and set to cool in a temperature under 50° C., after adding the foam of the whites of 5 beaten eggs. Finally, it is once more cooked for several minutes and then filtered. To the clear, straw-yellow filtrate 8- to 10-per-cent. glycerin is added, and after having been poured into the test-tubes they are sterilized in the steam-sterilizer for ten minutes on three successive days.

Lactic-acid Agar-agar and Wort Agar-agar.

The same as the preparation of lactic-acid gelatin, only instead of 10-per cent. gelatin 1- to 2-per-cent. agar-agar is added.

Nourishing Jelly, after Miquel; Melting-point, over 50° C. (9).

Three hundred to 400 grammes Irish moss (caragheen, *Fucus crispus*) are cooked for several hours in 1 litre of water, strained through a sieve and the strained fluid again cooked. It is then steamed over the water-bath, and then dried in a temperature of 40° to 45° C. One per cent. of this jelly renders bouillon firm at 45° to 50° C.

Blood-serum, according to Koch (10).

Blood is procured from a wounded animal which bleeds freely, and caught in a sterilized glass cylinder about 20 centimetres high and 8 to 10 centimetres in diameter, the mouth of which is closed by a glass capsule, and the whole allowed to remain undisturbed in an ice-box for two to twenty-four hours. The separated serum is then taken out by means of a pipette and placed in the sterilized test-tubes. By careful manipulation sterilization may be rendered unnecessary. If sterilization *is* necessary, it may be achieved by warming for one to two hours each day at 58° C. for five to six days. The serum so treated however, has a layer of

cholesterin upon its surface, which may often be mistaken for bacteria growth. The serum is hardened (desiccated) by placing it in a "desiccator" and warmed at 68° C. The serum so treated is hard and firm, amber-colored, and, if cows' or sheep's blood has been employed, transparent, or only in the deeper parts slightly milky. The water expressed by condensation is abundant

Blood-serum, after Loeffler (11).

To meat-broth are added 1-per-cent. peptone, 1-per-cent. grape-sugar, and 0.5-per-cent. table-salt, neutralize with natrium carbonate, cooked on the water-bath until the albuminous bodies are completely precipitated, and then filtered. This bouillon is sterilized in the steam-apparatus, and after it has cooled is mixed with fluid blood-serum in the proportions of 1 to 3; the whole is again sterilized by interrupted exposures in the steam-sterilizer at about 58° to 60° C., and then desiccated as in the preceding preparation.

Glycerin Blood-serum, according to Nocard and Roux (7).

To ordinary blood-serum add 6- to 8-per-cent. glycerin. Otherwise, the same as the preceding one, except that this must be heated to 75° to 78° C., in order to have the mixture perfect.

Blood-serum Gelatin.

Sterilized blood-serum is mixed with an equal mass of a concentrated solution of gelatin, the concentration of which is double the volume of the serum gelatin which is desired, procured by heating at 37° C. Afterward, the mixture may be heated several successive days at 52° C. for one to two hours.

Blood-serum Agar agar, after Hueppe (2).

Equal quantities of blood-serum, made alkaline by addition of a 2-per-cent. solution of carbonate of sodium, and liquefied 2-per-cent. agar-agar solution. This solution bears heat to the boiling-point, enabling us to use a high temperature in sterilization.

Desiccated Blood, after A. Pfeiffer (12).

Blood as procured under the same precautions as mentioned above, and poured into the test tubes (sterilized), and by a gradually-rising temperature brought to desiccation. Such a preparation is chocolate-brown colored, thoroughly opaque, and possesses a smooth, shining surface.

Alkali-albuminate, after Tarchanoff and Kolesinkoff (3).

(*a*) Bouillon Albuminate.—Hens' eggs, with their shells, are allowed to remain four days in a 5- to 10-per-cent. solution of kalium hydrate, whereby the albumen becomes gelatinous and transparent. They are then placed in water in the proportions of 10 to 100, and sterilized for three days in steam-sterilizer.

(*b*) Syrup-like Albuminate.—Chickens' eggs, with their shells, are soaked for four days in a 5- to 10-per cent. kalium-hydrate solution, the albumen thinned to one-half its solidity with water, and then sterilized in steam-cylinder.

(*c*) Solid Albuminate.—Chickens' eggs with their shells are laid in a 5- to 10-per-cent. solution of kalium hydrate for fourteen days, whereby the albumen becomes gelatinous, and, if torn, does not close together again; it is, however, transparent. This is cut into thin lamellæ, and manipulated the same as sliced potatoes. Sterilize in steam-sterilizer.

Rice-milk, after Kral (14).

One hundred grammes of rice-powder are thoroughly mixed with 250 cubic centimetres of skimmed milk, and after placing in a porcelain dish warmed over a Bunsen burner until it becomes colloidal; during this process it must be constantly agitated. While hot it is pressed firmly into a "potato-borer," by means of a horn spatula. After allowing to cool, the molded cylinder of rice-milk is shoved slightly forward out of the potato-borer, cut, by means of a

highly-strung bow of thin platinum wire, into pieces 6 to 7 millimetres in thickness, which are placed in glass boxes; then, after the addition of 8 drops of milk to each, they are placed in the steam-sterilizer for one to one and one-half hours.

Milk-bouillon Rice, after Soyka-Eisenberg.

One hundred grammes of rice-powder, 210 grammes milk, and 70 grammes bouillon are thoroughly triturated in a mortar, then placed in glass boxes, which are heated uncovered in a water-bath until it becomes stiff. After placing on the covers the boxes are sterilized for three successive days, for fifteen to twenty minutes. The mass is then homogeneous, dirty-white or milk-and-coffee colored, with a smooth surface.

Potatoes.

(*a*) Unpeeled Cut Potatoes, after Koch (1).—Unpeeled potatoes are thoroughly cleansed with a brush, then washed in 1-per-cent. sublimate solution, and then allowed to soak in 1-per-cent. sublimate solution for one-half to one hour; they are next thoroughly cleansed in water, after which they are cooked in the steam-sterilizer for three-fourths to one hour. The potatoes are now ready to be cut in half. The hands are first sterilized, and the knife to be used heated to red heat. After cutting they are placed in damp chambers to await employment.

(*b*) Peeled Potatoes, after Esmarch (15).—The potatoes are peeled, washed, the "eyes" and rotten spots cut away, and then cut into pieces about 1 centimetre thick; these are placed in double-saucers. Sterilize by exposure in the steam-sterilizer for three-fourths to one hour on three successive days. Soyka's (16) modification consists in the use of peculiar dishes.

(*c*) Cylinders of Peeled Potatoes, after Bolton (17), Globig (18).—By means of an "apple-corer" or a large cork-cutter cylinders are cut from peeled potatoes; the diameter of such cylinders must be smaller than the test-tubes into which they are to be placed. After cutting they are sliced diagonally in half, and each half placed in a test-tube with the large, slanting surface uppermost. Sterilize in the test-tubes for three successive days. The modifications of Roux (19), Hueppe (2), Gunther (20), consist in peculiar appliances for retaining the moisture of the potatoes.

(*d*) Potato-Meal.—Peeled potatoes are cooked for three-fourths of an hour in the steam-sterilizer and pressed into Erlenmayer's bulbs. Eisenberg's modification (21): The potato-meal is pressed with a spatula into glass dishes, having plain, unrimmed glass covers. Sterilize as usual. Seal with paraffin for lasting culture.

Potatoes Cut into Disks, after Wood (2).

From good, white potatoes disks are cut, which are pressed on strips of glass which are placed in test-tubes. Sterilize by steam.

Bread-pap.

Dry, gray bread is rubbed into a powder, with which Erlenmayer's bulbs are filled to ½ centimetre, and with a little water brought to a pap. Neutralize by means of natrium carbonate if it is to be used for bacteria; not if for fungi. Neutralize in steam.

Fresh Egg for Anaerobic Cultures, after Hueppe (22).

Fresh eggs are thoroughly cleaned and sterilized by means of sublimate solution, rinsed in sterilized water, and dried in sterilized cotton. The inoculation of such an egg is procured through a fine hole in the shell made by a glowing needle, which, after inoculation, is closed by means of a small piece of fine, sterilized paper, over which a layer of collodium is laid.

Meat-disks, after Kral (14).

Fresh meat is carefully freed from connective tissue, chopped fine, and then spread out thinly upon large glass plates, and dried in a sterile air at 40° to 50° C. The horny mass which results is rubbed into a fine powder and sifted through a fine sieve; 100 grammes of

this meat-powder are mixed with 300 cubic centimetres of peptonized bouillon and rubbed up in a porcelain dish to a pap. This is placed upon circular glass disks, which have been moistened with glycerin, and the disks placed one upon the other (10 to 15). They are then placed in a sheat-iron box filled with bouillon and left fifteen minutes in a temperature of 100° C. The glass disks are then carefully separated, the meat-layers detached and shaped with the cork-cutter, and then placed in glass dishes; after which they are sterilized by exposure for one to one and one-half hours in a stream of steam.

Wafers, after Schill (23).

Ordinary church-wafers are moistened in a nourishing media and sterilized in glass dishes. They make good media, especially for chromogenic bacteria, the products of which are made prominent by the white background.

USE OF THE NOURISHING MEDIA.

Test-tube Cultures.

Ten cubic centimetres of the nourishing media in a fluid condition are poured into the test-tubes, the circumference of which is not too small nor the glass very thin. Care should be exercised, in pouring the media into the tubes, so that the mouths of the tubes are quite free from the same, otherwise much annoyance may be occasioned by the media hardening around the cotton plugs, thus rendering their withdrawal difficult, and that at a time when all manipulation should be performed with rapidity and dexterity. The tubes are closed with a firm cotton plug. It is not absolutely necessary to sterilize the empty tubes in dry air before pouring in the media, as the subsequent exposure of them in the steam-sterilizer is sufficient for practical purposes; but new test-tubes should be first thoroughly washed in acidulated water, inasmuch as these have usually a strong alkaline reaction, and this, in the course of the subsequent boiling of the gelatin, renders the media cloudy. During the process of solidification the tubes containing gelatin are usually allowed to stand upright, so that the surface of the gelatin is at right angles with the sides of the tube; with tubes containing serum or agar-agar, they are placed in an oblique position, so that the media has a sloping and therefore the greatest area of surface. The cultures upon the former are called stab cultures; those upon the latter, stroke cultures.

Object-glass Cultures, after Koch.

Fluidified gelatin is poured upon the surface of a sterilized object-glass, allowed to solidify, and then inoculated by stroking the surface with the infected platinum needle.

Plate Cultures, after Koch.

These are used in isolating the various bacteria and in procuring pure cultures of the same, and may be employed with all the gelatinous and agar media, as well as with serum. A tube of liquefied media is first inoculated and thoroughly mixed with the material to be examined. In pathological fluids, 3 to 4 platinum ösen are required; of water, beer, milk, etc., 1 cubic centimetre. Then follows the process of attenuation, thus: From the first tube 3 ösen are taken and carried over to the second tube of liquefied gelatin, and from this second tube 3 ösen are carried over to a third tube (water requires $\frac{1}{2}$ cubic centimetre from the first to second tube, etc.). After thorough mixture and sterilization of the mouths of the tubes by heating in the gas-flame, their contents are poured evenly upon the surface of the sterilized glass plates, which are then placed upon the leveling apparatus and allowed to cool; after which they are ready for use in the damp chamber.

Modifications, after Schimmelbusch (24), *Schill* (23), *Babes* (26), *Patri* (27), *Soyka* (28).

These modifications have for their object: to avoid the necessity of the leveling apparatus, to exclude as far as possible the contamination from the air, and the prevention of

drying of the nourishing media, especially agar agar, by retaining the condensation water. They consist, in part, of two glass plates separated by a piece of dry pasteboard, and held together by a metal clamp, or of flat flasks with parallel walls and an opening at the side, or of flat, glass double-dishes.

Bulb Cultures, after Salomonsen (29), *Cramer* (30), *Cornil-Babes* (31), *Kowalsky* (32).

For the prevention of air infection the nourishing media is placed in sterilized Ehrlenmeyer's bulbs, which are stoppered with a cotton plug. Pasteur's bulbs are sealed by fusing the glass. The disadvantage of all these methods is, first, that the culture cannot be placed under the microscope, but can only be examined by the hand magnifying glass, and, second, the material contained in the bulbs is very difficult to get at.

Roll Culture, after Esmarch (33).

Gelatin or agar (with the addition of 1- to 2-per-cent. gum arabic) is liquefied in the test-tubes, inoculated as before described, and the cotton plug covered by a rubber cap. The media is then made to spread upon the sides of the tubes in an even layer by turning the tubes around and around in a dish of ice-cold water. This method has many points of advantage,—rapidity of results with the minimum amount of apparatus, prevention of air infection, and unusual length of time the growth may be observed. The only disadvantage is that those colonies which liquefy gelatin soon sink to the bottom of the tube, thereby preventing further observation.

Methods for Aerobic Bacteria.

By Atmospheric Restriction.

After Koch (34), by laying thin sheets of mica on the gelatin or agar at the moment of hardening.

After Hesse (35), by pouring gelatin or sterilized oil on tube culture.

After Esmarch (33), by pouring fluid gelatin into the centre of roll culture, and hardening by at once placing the tube in ice-water.

After Buchner (36), by placing ordinary tube culture and roll culture in a larger tube which contains 1 gramme of pyrogallic acid, to which is added 10 cubic centimetres of $\frac{1}{10}$ solution of kalium, and which is then hermetically sealed. The alkaline pyrogallol absorbs the oxygen; so that the culture is contained in an atmosphere of hydrogen, with traces of carbonic acid and carbonic oxide.

After Hueppe (2), in fresh eggs. (See Hueppe's method.)

After Kitasato-Weyl (37), by the addition of some active reducing agent to the solid nourishing media. The best is 0.3- to 0.5-per cent. formic acid to liquefied agar-agar. Sterilize as usual.

After Fuchs (38). The condensation water is first poured off from the inoculated tube. The tube is then inverted, and by means of glass tubing hydrogen is led into it for several minutes; after which it is closed by a rubber cap and sealed with paraffin.

Complete Exclusion of Air.

Under Hydrogen, after Buchner (39), *Hauser* (40), *Liborius* (41).

At the side of the test-tube is a protuberance for the purpose of connecting the interior of the tube with the gas-apparatus. After inoculation the tubes are melted together beneath the cotton plugs, while the hydrogen-gas is still entering. Afterward the protuberances at the sides of the tubes are also sealed together by heat.

In Hydrogen-gas, after Roux (42) *and Liborius* (41).

These éprouvettes have a right-angle tube which extends to the bottom of the same internally. Thus, after inoculation the gas is conducted through the liquefied media for about ten minutes. The tubes are then melted together.

In Hydrogen, after Hueppe (2) *and C. Fraenkel* (13).

The éprouvettes are closed by rubber corks, which have two holes. Through one of these a long tube passes to the bottom of the media, and to which communication is procured with the gas apparatus; the other shorter tube, which ends immediately below the cork, is for the purpose of conducting the gas off. The gas is allowed to pass through the media after inoculation for about ten to twenty minutes, when the egress tube is sealed, and then the ingress tube. In the case of agar being employed, the tube must be kept at a temperature of 40° to 42° C.; gelatin, 30° to 35° C. The shape of the Fraenkel tube renders plate methods possible.

Substitution of Air by Steam—"High Culture," after Lœbrius (41).

An éprouvette, after being filled with the media, is, by means of heating over a flame or immersion in simmering water, freed from air, and then quickly plunged into cold water (temperature 30° to 40° C.) and allowed to cool. It is then inoculated, mixed, and the glass melted together.

Exhaustion of Air by Means of Air-pump, after M. Gruber (44).

Tubes larger in length, breadth, and thickness than the ordinary tubes are required. About 5 centimetres below their mouths they have a constriction. The tubes are filled with about 10 cubic centimetres of nourishing media, stoppered with cotton, and sterilized. When inoculated, the cotton plug is pushed deep into the tube to the narrowed portion and a rubber cork inserted, through which passes a piece of tubing bent at right angles. While the tube is being heated at 30° to 35° C., or if agar-agar at 40° to 47° C, in the water-bath, the tubing is connected with an air-pump and the air evacuated from the tube. To prevent foaming of the contents the tube may be gently heated over a Bunsen burner just below the constricted portion. The tubes are then melted together below the constriction, and the media solidified according to Esmarch's roll method. In a like manner we may observe the anaerobic bacteria growing on potatoes.

STAINS AND REAGENTS.

Staining Solutions.

Concentrated Alcoholic Solution.

The coloring agent, in powder, is placed in a bottle and absolute alcohol poured upon it; the whole is then well shaken and allowed to stand. After a time a certain amount of undissolved coloring matter must have fallen to the bottom: otherwise, more is added. As the solution is consumed, fresh alcohol is added.

Aqueous Solutions and Dilute Alcoholic Solutions.

To distilled water add concentrated alcoholic solution sufficient to color it deeply, but the bottom of the watch-glass must always be visible. The stains usually thus employed are: fuchsin, gentian violet, methyl-violet, Bismarck brown, or vesuvin; the latter may be kept in concentrated solution, in equal parts of glycerin and water, after Koch. The stains most durable and having the greatest staining-power are gentian violet, fuchsin, and, laterally, Bismarck brown. Methylene blue stains weaklier and more slowly, and is rather permanent, but hardly ever overstains.

Alkaline Solution of Methylene Blue.

(*a*) Weak, after Koch (15).

Concentrated alcoholic solution of methylene blue,	1.0 cubic centimetre.
Distilled water, ...	200.0 cubic centimetres.
Kalium solution (10 per cent.), ...	0.2 cubic centimetre.

(*b*) Strong, after Loeffler (11).

Concentrated alcoholic solution of methylene blue,	30.0 cubic centimetres.
Kalium solution (0.01 per cent.), . . .	100.0 cubic centimetres.

(*c*) After Schütz (16).

Concentrated alcoholic solution of methylene blue,
Kalium solution (0.01 per cent.), ... ää equal parts.

Aniline-water Solution.

(*a*) Aniline oil is added to distilled water until oversaturated, well agitated, and allowed to stand for several minutes; after which it is filtered, the filter-paper having been previously moistened with water. To the thoroughly clear filtrate (aniline water) enough of a concentrated solution of fuchsin, or methylene violet, is added to produce opalescence.

(*b*) Modification after Weigert-Koch (15).

Saturated aniline water, ...	100 cubic centimetres.
Concentrated alcoholic solution of methylene blue, or fuchsin, ...	11 cubic centimetres.
Absolute alcohol, ...	10 cubic centimetres.

The solution will keep ten to twelve days.

Weak Alkaline Aniline-water Solution, after Loeffler (18).

To 100 cubic centimetres of saturated aniline water 1 cubic centimetre of a 1-per-cent. solution of NaHO is added. Fresh aniline water has a neutral reaction, and after the addition of the sodium solution it has a marked alkalinity. This alkaline aniline-water is poured into an Erlenmeyer bulb, in which has been placed 4 to 5 grammes of methylene blue, methylene violet, or fuchsin. After stopping the mouth of the bulb with a good rubber cork, it is well shaken. We now have a very concentrated solution of the stain, which keeps well for a week. In using, a few drops are filtered off on the cover-glass.

Carbolic-acid Solution.

(*a*) After Ziehl Neelsen (49).

Distilled water, ...	100 grammes.
Acid. carbolic. cryst., ...	5 grammes.
Alcohol, ...	10 grammes.
Fuchsin, ...	1 gramme.

This solution is permanent.

(*b*) After Kuehne (50).

Methylene blue, ...	1.5 grammes.
Alcohol (absolute), ...	10. grammes.

This is rubbed up gradually with 100 cubic centimetres of 5-per-cent. carbolic-acid solution. The staining-power of this solution is increased with time.

Ammonia Solution, after Weigert (51).

Liq. ammon., ...	0.5 gramme.
Alcohol. absolut..	10.0 grammes.
Aq. destil.	90.0 grammes.
Gentian violet, ...	2.0 grammes.

Ammonia carbolate Solution, after M. Hermann (52).

(*a*) Crystal violet, ...	1 gramme.
Alcohol (95 per cent.), ...	30 c.cm.
(*b*) Ammon. carb., ...	1 gramme.
Distilled water, ...	100 c.cm.

Enough of solution *a* is added to solution *b* to produce on filter-paper a dense stain.

Borax Solution, after Sahli (53).

Distilled water, ...	40.0 grammes.
Sat. aqueous sol. methylene blue, ...	24.0 grammes.
Borax sol. (5 per cent.), ...	16.0 grammes.

Borfuchsin, after Luebimoff (54).

To 20 grammes distilled water 0.5 gramme of boric acid is added, after which 15.0 grammes absolute alcohol, whereupon the crystals are nearly all dissolved. Lastly, 0.5 grammes fuchsin is added and made to dissolve by agitating the flask. The completed slightly-acid solution is transparent and clear.

Aniline-oil and Clove-oil Solution, after Kuehne (50).

As much of the stain (methylene blue, safranin, methyl-green, auramin, acid violet, fluorescin) as may be held on the point of a knife blade is rubbed lightly in a mortar with 10.0 grammes of clarified aniline oil or 15.0 grammes clove-oil. If the coloring-matter is not entirely dissolved the whole is poured unfiltered into a flask and allowed to stand until the undissolved stain has been deposited and the oil is transparent. Several drops of this are removed when required and added to pure oil until the desired concentration is produced.

Acetic-acid Gentian-violet Solution, after Friedlaender (57).

Concentrated alcoholic solution of gentian violet,	50 grammes.
Acetic acid,	10 grammes.
Distilled water,	100 grammes.

Carmine Solutions (50).

(*a*) After Cuccati. Twenty grammes crystallized carbonate of sodium are dissolved in 100 grammes warm water, 50 grammes carmine added, boiled, taken from the fire, and 30 grammes absolute alcohol added. Several days after the solution is filtered, and the following ingredients are slowly added: 300 grammes of water, 8 grammes of a 20-per-cent. solution of acetic acid, 2 grammes chloral hydrate. Time requisite for staining about one-fourth hour.

(*b*) Lithion Carmine, after Orth. To a cold saturated aqueous solution of carbonate of lithium 25-per-cent. carmine is added, cooked for ten minutes, and, after cooling, filtered.

(*c*) Hydrochloric-acid Carmine. To 50 grammes alcohol (60 to 80 per cent.) add 4 drops of hydrochloric acid and 0.5 gramme carmine, cook for ten minutes, and, after cooling, filtered.

Delafield's Hæmatoxylin Solution (50).

To 200 cubic centimetres of a concentrated aqueous solution of ammonia a solution of 2 grammes hæmatoxylin in 12 cubic centimetres of absolute acid is added. After this mixture has stood exposed to air and light for three to four days it is filtered and mixed with 50 grammes glycerin and 50 grammes methyl-alcohol. The solution is allowed to stand until it assumes a dark color, whereupon it is again filtered and stored in a glass stoppered bottle. When needed, the solution is diluted with more or less water, according to the rapidity of the stain. The weak solution gives the best results.

Fluorescin-alcohol (50).

One gramme fluorescin is rubbed up with 50 cubic centimetres absolute alcohol and the mixture poured into a bottle. After a time the undissolved fluorescin is deposited. When about half of the clear solution is used up, fresh alcohol is poured in, and so on until the undissolved fluorescin is present.

Iodide-of-kalium Solution.

(*a*) After Gram.

Iodine,	1 gramme.
Iodide of kalium,	20 grammes.
Distilled water,	300 grammes.

(*b*) After Kuehne.

Iodine,	2 grammes.
Iodide of kalium,	4 grammes.
Distilled water,	100 grammes.

When required, enough of either of these solutions is added to water until it assumes the color of madeira.

Acid Solutions for Decolorization.

Sulphuric, hydrochloric, or nitric acid, in about 25-per-cent. aqueous solution.
Acetic acid, in ½- to 1-per-cent. solution.

Acid Alcohol.

Nitric acid,	1 part.
Alcohol,	10 parts.

Macerating Fluid, after Loeffler (59).

To 10 cubic centimetres of a 20 per cent. aqueous solution of tannin add, drop by drop, an aqueous solution of sulphate of iron until the solution assumes a deep-violet color. To this add 3 or 4 cubic centimetres of logwood infusion (1 part wood to 8 parts water). The addition of a greater quantity of this logwood infusion produces a granular condition, which renders the staining functions of the solution useless. The fluid assumes a dark-violet tone, which it retains for several days, and then it gradually changes and takes on a deep-black color. At the same time a thin skum develops upon the surface, and on the sides of the vessels containing it dark granular bodies are deposited; this does not interfere with the usefulness of the solution, which should be stored in well-fitting glass-stoppered bottles. The addition of 4 or 5 cubic centimetres of a 5-per-cent. solution of carbolic acid renders the solution permanent and does not affect its macerating strength materially.

Macerating Fluid, after Treukmann (60).

(*a*) Tannin,	1 per cent.
Hydrochloric acid,	½ per cent.
(*b*) Concentrated solution extract logwood.	
Hydrochloric acid,	½ per cent.; or
Gallic acid,	½ per cent.; or
Carbolic acid,	1 to 2 per cent.
(*c*) Saturated solution of catechu acid (produced by soaking catechu powder in excess of water for several days and filtering),	4 parts.
Saturated aqueous solution of carbolic acid,	1 part.

The same, after Loeffler (61).

Tannin solution (20 to 80 water),	10 c.cm.
Cold saturated solution of sulphate of iron,	5 c.cm.
Aqueous or alcoholic solution of fuchsin, methyl-violet, or wool-black,	1 c.cm.

The suitable fixing agent for individual organisms, whether 1-per-cent. solution of sodium or any of the acid solutions herein given, must be ascertained by several trials.

MICROSCOPIC EXAMINATION OF THE BACTERIA.

Detection of Bacteria in Fluids (Nourishing Media, Pus, Blood, Sputum, Water, etc.).

(*a*) Uncolored, on Glass Slide. A cover-glass is thoroughly cleaned and passed several times through the gas-flame. A drop of the fluid to be examined is quickly carried over on a platinum needle to the cover-glass, which is as quickly inverted over a concavity in an object-glass and made fast by means of vaselin. With the assistance of the color glass in the sub-stage the form and motility of the various bacteria may then be examined.

(*b*) Stained Cover-glass Preparations. The Koch-Loeffler process is to place a drop of the fluid on a clean cover-glass, with or without the addition of a drop of sterilized water, and evenly spread it over the surface of a cover-glass, by means of the platinum needle, or, in the case of thick fluids, by placing another clean cover-glass in contact with the first and drawing it across the same in a parallel direction until the fluid is evenly distributed, and then drying it in the air. It is then carried thrice through the flame and stained as follows:—

Simple Staining.

The cover-glass, prepared as aforesaid, is held in a pincette by one corner and a drop of the stain (concentrated aqueous basic aniline solution) is brought in contact with it and allowed so to remain one to five minutes, when the excess of stain is rinsed off with distilled water. Examine in the same, or, if it is to be preserved, dry in the air and mount in Canada balsam. Gunther rinses the preparation in 1- to 5-per-cent. acetic-acid solution previous to staining.

Isolating Staining of Bacteria by Decolorization, after Gram (62).

The preparation, stained with the aniline-water gentian violet of Ehrlich, is placed for one minute in the iodide-of-kalium solution, and then immediately in absolute alcohol, until the preparation is colorless. Rinse in water. The decolorized elements may be stained with Bismarck-brown solution for one minute.

Capsule Staining.

(*a*) After Ribbert (63). The cover glass preparation is hastily exposed in Ribbert's solution and immediately rinsed in water. The bacilli are dark, the capsules light blue.

(*b*) After Friedlaender. Stain for twenty-four hours in Friedlaender's acetic-acid gentian-violet solution, decolorize in 0.1-per-cent. acetic-acid solution, and rinse in water.

Spore Staining.

(*a*) After Neisser-Bienstock (61). Stain with heated aniline-water fuchsin, wash in hydrochloric-acid alcohol, double stain with methylene blue. Spores red, bacteria blue.

(*b*) After Buchner-Hueppe (65, 66). The cover-glass, prepared after Koch-Loeffler, is exposed to dry heat at 210° C. for one-half to one hour, or in moist heat at 120° C. for one hour, or touched with concentrated English sulphuric acid, and, after fifteen seconds, carefully washed according to Hueppe (thrice) and drawn through the flame seven to ten times. The staining then proceeds according to that employed with the tubercle bacillus by Ehrlich and Rindfleisch.

(*c*) After Hauser (67). A cover-glass preparation, after having been thrice passed through the flame, is covered with a concentrated aqueous solution of fuchsin, and again passed through the flame thirty to forty times until steam begins to arise from it. It is then exposed to 25-per-cent. sulphuric acid for several seconds, washed in water, and double stained in aqueous solution of methylene-blue solution.

(*d*) After Ernst (68). The cover-glass preparation, while yet warm from being passed thrice through the flame, is covered with as much as possible of the strongly alkaline methylene-blue solution of Loeffler. It is then picked up in a pair of pincettes by one of its corners and moved over the blue flame of a Bunsen burner until steam arises from it, care being taken that it does not boil. Rinse in water and double stain in Bismarck-brown solution for one to two minutes, or in very much diluted fuchsin solution. Spores are stained blue.

(*e*) After Neisser (69). Stain in heated carbol-fuchsin, rapidly rinse in 1-per-cent. aqueous solution of sulphuric acid, double stain in aqueous or Loeffler's solution of methylene blue: or, stain in aniline-water methylene-violet solution, wash in 1-per-cent. aqueous solution of sulphuric acid, and after-stain in acid brown.

Staining of Special Parts of Bacteria, after Babes (70).

Concentrated Loeffler's methylene blue is dropped on a cover-glass which has been only *partially* dried and allowed to remain for about one-fourth of an hour until the stain is beginning to dry up; it is then carefully rinsed in water, and mounted in water or Canada balsam.

Bacteria Nuclear Stain, after Ernst (71).

(*a*) Stain with warm, not hot, alkaline methylene-blue solution, wash in water, and after-stain in cold Bismarck-brown solution. The nuclei (sporogenous spots) are blue-black, thus differentiating the light-blue-stained spores. This effect is produced by "mixed staining."

(*b*) Stain with Delafield's hæmatoxylin; the bacillus is of a light-blue color, the nuclei dark violet.

(*c*) Stain with Plattner's nucleus-black; the nuclei are black.

Flagella Staining.

(*a*) After Koch (72). Stain with a concentrated solution of extract of logwood; then immerse in 0.5-per-cent. chromic acid solution, or in Muller's solution. Under this condition, a brown-black insoluble combination of the extract of logwood with the chromic acid is formed. Rinse in water, dry, and mount.

(*b*) After Künstler (73). Several drops of the fluid to be examined is placed on the object-glass and a small drop of osmic acid added to it. It is allowed to stand for fifteen minutes to evaporate. It is then covered with a cover-glass, and a drop of concentrated Collins black placed in the middle of each of the four sides, which is made to mix with the stain under the glass by teasing with a needle. The glass is then surrounded by paraffin and then with wax to prevent further evaporation. In about eight to fourteen days the cilia are visibly stained.

(*c*) After Neuhaus (74). The prepared cover-glass is allowed to swim on the surface of a warm solution of sulphate of iron for a few minutes; then stain in logwood solution, after which soak in a neutral solution of chromate of sodium. This latter solution is produced by adding 5-per-cent. sodium solution, drop by drop, to a weak solution of chromic acid. Or:—

Boil the cover-glass five minutes in the common "*Kaiser tint*," then soak for fifteen minutes in a very weak solution of neutral chromic-acid sodium which has been warmed. This process is repeated twice or thrice.

(*d*) After Trenkmann (60). A very small drop of the fluid to be examined is placed on the cover-glass, to which is then added a large drop of distilled water, and the whole is spread evenly over the surface of the glass and allowed to dry in the air without heating. Then:—

1. Two to twelve hours in the tannin-beize, wash in a weak solution of dahlia (2 drops concentrated alcoholic solution of dahlia to 20.0 water), or fuchsin (2 to 4 drops concentrated alcoholic solution of fuchsin in 20.0 water), or gentian violet (1 drop in 80.0 water), or carbol fuchsin (2 drops to 20.0 of 1-per-cent. carbolic-acid solution. After one to four hours in the stain, wash in water.

2. Two to twelve hours in concentrated solution of extract of logwood Wash; stain in some acid aniline stain.

3. Two to twelve hours in catechu acid. Wash; stain in some aniline stain.

(*e*) After Loeffler (61). The cover-glass must always be perfectly clean and free from grease. To this end they should be treated as follows: Immerse in concentrated sulphuric acid and warm, rinse in distilled water, and then rub dry with a cloth free from grease after soaking in a solution composed of equal parts alcohol and ammonia. A drop of the fluid to be examined is then placed on the cover-glass by means of the platinum needle, several drops of water added, and the whole carefully spread out over the surface of the glass. Dry in the air, and afterward pass three times through the gas-flame, being careful that it is not heated too highly. A few drops of beize are then placed upon the cover-glass while it is yet warm, whereupon it is again moved to and fro over the flame until steam begins to arise, and, after a half to one minute, is washed in distilled water, then in absolute alcohol, until the cover-glass appears completely colorless. This operation is repeated entire, and the preparation is then ready to be examined. The best stain is a neutral, saturated solution of aniline-water fuchsin.

Gonococcus Staining.

(*a*) After Neisser (75). The preparation is first floated on the surface of a concentrated alcoholic solution of eosin, which is then heated. The excess of eosin is then removed from the cover-glass by means of blotting-paper. It is then placed in a concentrated alcoholic solution of methylene blue for one-fourth of a minute. Rinse in water.

(*b*) After Schütz (76). Stain for five to ten minutes in a cold saturated solution of methylene blue in 5-per-cent. carbol-water, which has been filtered; wash with water; dip in acetic-acid water (5 drops of dilute acetic acid in 20 cubic centimetres distilled water). Double stain with very much diluted solution of safranin. Gonococcus, blue; pus-cells and their nuclei, salmon-colored.

(*c*) After Steinschneider and Galewski. To differentiate from other diplococci, preparation is placed in aniline gentian violet for twenty-five to thirty minutes, washed; then in iodide-of-potassium solution for one or five minutes; then in alcohol until the preparation is decolorized. After again washing and drying, after-stain for several seconds in Loeffler's methylene-blue solution; the gonococci are then lightly and the other diplococci darkly stained.

Tubercle-Bacillus Staining.

(*a*) After Koch and Ehrlich (77). Cover-glass preparations, dried and fixed in the air, are stained for twelve hours in Weigert-Koch aniline-water aniline-stain solution, then dipped for one to three seconds in weakened sulphuric acid (1 to 3 or 4), and at once agitated in 60-per-cent. alcohol. After-stain in aqueous solution of vesuvin or methylene blue for several minutes.

(*b*) After Rindfleisch (78). The staining solution is exposed to heat until steam arises and bubbles appear on its surface. Otherwise, the same as above.

(*c*) After B. Fränkel (55). Boil aniline water in a test-tube, pour into watch-glass, add as many drops of a concentrated alcoholic solution of fuchsin as will produce a shimmering color to the surface of same. The cover-glass preparations are then allowed to swim on the surface of this solution for five to ten minutes, after which it is placed in Fränkel's methylene-blue sulphuric-acid solution for one to two minutes. Rinse in water or ½-per-cent. acetic-acid water.

(*d*) After Ziehl and Neelsen (19). The cover-glass preparation is stained in boiling Ziehl-Neelsen carbol-fuchsin solution. After a short time rinse in water and decolorize for a moment in 5-per-cent. sulphuric acid or diluted nitric acid (1 to 3), then in 70-per-cent. alcohol and water. After-stain in methylene blue.

(*e*) After Weichselbaum (79). Stain as with Ziehl-Neelsen, and then, after rinsing in water, directly in a concentrated alcoholic solution of methylene blue, where the cover-glass is allowed to remain until it is evenly stained blue. Rinse in water.

(*f*) After Kaatzer (80). The cover-glass preparation is placed in an oversaturated alcoholic solution of gentian violet for twenty-four hours, or, if the solution is warmed to 80° C., for three minutes. Decolorize in a solution composed of 100 cubic centimetres 90-per-cent. alcohol, 20 cubic centimetres water, and 20 drops concentrated hydrochloric acid. Wash in 90-per-cent. alcohol. After-stain with concentrated aqueous solution vesuvin solution.

(*g*) Pfuhl-Petri (81). Stain in a warmed solution composed of 10 cubic centimetres of saturated alcoholic fuchsin solution to 100 cubic centimetres water for one to two minutes. Decolorize in acetic acid for one to two minutes, rinse in water, and double stain in alcoholic aqueous solution of malachite green for one-half to one minute; rinse in water, dry, and mount in balsam.

(*h*) After Lübimoff (54). Stain with borax-fuchsin as in Koch Ehrlich method. Decolorize in sulphuric acid (1 to 5).

(*i*) After M. Hermann (52). In the heated mixture of Hermann's solution *a* and *b*, the cover-glass preparation is allowed to remain not to exceed one minute, then for four to five seconds in $\frac{1}{10}$-per-cent. nitric acid. Wash in 95-per-cent. alcohol, and after-stain in eosin (1 gramme to 100 cubic centimetres 60-per-cent. alcohol) one-half minute.

(*k*) After Gabett-Ernst (56). Stain cover-glass preparation in cold Ziehl-Neelsen's solution for two to five minutes, then for one minute in Gabett's methylene-blue sulphuric-acid solution, and rinse in water. This method is the quickest, most convenient, and most exact.

The methods of Gram, Kuehne, Weigert, Lustgarten, which, though not specific for the tubercle bacillus, yet stain the same, will be spoken of under "Staining of Sections."

Lepra-Bacillus Staining.

(*a*) After Baumgarten (82). Stain for six to seven minutes in dilute alcoholic solution of fuchsin, and decolorize for one-fourth minute in acidulated alcohol (nitric acid and alcohol 1 to 10); rinse in water, double stain in aqueous solution of methylene blue. Lepra bacilli red on blue ground. Tubercle bacilli do not stain in this space of time.

(*b*) After Lustgarten (83). Stain with aniline-water fuchsin or gentian violet, and decolorize in 1-per-cent. chloridate of sodium for a long time, then rinse thoroughly in water. The tubercle bacilli are decolorized earlier than the lepra bacilli.

Syphilis-Bacillus Staining.

(*a*) After Lustgarten (83). Stain in aniline-water gentian violet for twenty-four hours, rinse in water, place in 1½-per-cent. solution of kalium permanganate for ten seconds, then for one or two seconds in an aqueous solution of sulphurous acid (produced by treating metallic copper with sulphuric acid), and rinse in water. If the preparation is insufficiently stained, the treatment with kalium permanganate and sulphurous acid is repeated for three or four seconds, until complete staining is accomplished. A durable stain is impossible.

(*b*) After Giacomi (84). Stain for several minutes in aniline-water fuchsin, then place in water to which has been added several drops of chloride-of-iron solution, and finally rinse in water. The syphilis bacilli lose their color in mineral acids at once, or very quickly,—thirty-five to forty-five seconds (differentiation between tubercle and leprosy bacillus),—but bear treatment in alcohol for a longer time without effect (differentiation between smegma bacillus: Alvarez and Tavel).

(*c*) After J. Lewy (85). Stain with carbol-fuchsin and decolorize with water.

DETECTION OF BACTERIA IN TISSUE.

Hardening.

1. In Alcohol. Small pieces of tissue, about the size of a hazel-nut, are taken while warm from the body, when possible, and placed in a bottle containing absolute alcohol, and having at its bottom a piece of filter-paper. Care must be taken that fresh absolute alcohol be added to the alcohol as it takes up the water of the tissue and of the air. In about two days the tissue is hardened.

2. In Chromic Acid. Place the piece of tissue in 0.1- to 0.5-per-cent. solution, and allow to remain one to eight days. Afterward, rinse in water until the water is not discolored. Then place in 90-per-cent. alcohol, then in absolute.

3. In Picric Acid. The piece of tissue remains for one to two days in concentrated aqueous solution of picric acid, is then washed in running water for twenty-four hours, and then carried gradually through 50-per-cent. to absolute alcohol.

4. In Sublimate. The tissue is taken as warm as possible and placed in 5-per-cent. corrosive-sublimate solution, which is warmed to 60° or 70° C. for about ten to thirty minutes. Then, into 70- to 80-per-cent. alcohol for one day; from this into 90-per-cent., and then in absolute alcohol.

Fastening and Cutting of Tissue.

The piece of tissue is fastened to a small cork by means of glycerin-gelatin: 1 part gelatin, 2 parts water, 4 parts glycerin, made into a solution by warming, and then cooked for a short time; to hinder the development of bacteria in the same, it is well to add a few drops of sublimate solution. A drop of this glycerin-gelatin is placed on a cork, and the piece of tissue pressed into it; then it is exposed to the air for about one minute, and then the whole is replaced in alcohol. After a few hours it is ready for the microtome. Before coloring, always place the sections in alcohol.

Staining of Sections—Common Methods.

1. After Loeffler. The section is placed in Loeffler's methylene-blue solution (or the concentrated aqueous solution of any stain will do equally as well); then for a few seconds in 0.5-per-cent. acetic-acid solution, absolute alcohol, cedar-oil; then Canada balsam.

2. After Koch. The stained section is placed for five minutes in a saturated solution of carbonate of kalium, prepared by mixing a saturated solution of the salt with equal parts of distilled water. Then in alcohol to dehydrate; then cedar-oil; then balsam.

3. After Schütz (86.) Stain in kalium solution, 1 to 10,000, and concentrated alcoholic methylene-blue solution, equal parts, for about twenty-four hours. Rinse in water which contains 4 drops of acetic acid; alcohol (50 per cent.) for five minutes; alcohol, absolute, fifteen minutes; cedar-oil and Canada balsam.

4. After Gram. Stain in aniline-water gentian violet one to three minutes. Rinse in water; iodide-of-kalium solution two and one-half to three minutes. Repeated washings in 90-per-cent. alcohol; absolute alcohol, cedar-oil, and Canada balsam.

5. After Günther, Modification of Gram's Method (87). Stain the section in aniline-water gentian violet one minute. Dry with filter-paper; iodide-of-kalium solution two minutes; alcohol one-half minute; hydrochloric acid, 3 per cent., ten seconds; alcohol, cedar-oil, and Canada balsam.

6. Kuehne's Modification of Gram's Method (50): (*a*) Stain the section in a solution composed of 1 gramme Victoria blue, in 50 cubic centimetres 50-per-cent. alcohol, added to an equal part of a 1-per-cent. aqueous solution of carbonate of ammonia for five minutes. Rinse in water; kalium iodide solution (2 : 4 : 100) two to three minutes. Rinse in water; extraction of stain by fluorescin alcohol. Rinse in pure alcohol; carry over into aniline oil; extraction of aniline oil by immersion in a high ethereal oil and xylol; Canada balsam. (*b*) Stain the section in concentrated aqueous solution of violet, to which has been added hydrochloric-acid (1 drop to 50 drops of water). Rinse in water; iodide-of-kalium solution two to three minutes. Rinse in water; absolute alcohol for several seconds; extract in aniline oil; ethereal oil and xylol; balsam.

7. Kuehne's Carbol-Methylene-Blue Method (50). Stain in carbol-methylene blue one-half to two hours. Rinse in water. Extract in acidulated water (10 drops HCl, 100 drops water). Rinse in a weak aqueous solution of carbonate of lithium (10 drops water, 6 to 8 drops of a saturated solution carbonate of lithium): then in pure water; dip in absolute alcohol containing a small quantity of methylene blue; methylene-blue aniline oil a few minutes. Clear in pure aniline oil; ethereal oil (cloves, terpine) two minutes. Extract oil in xylol; balsam.

8. Kuehne's Fuchsin Method (50). Stain in carbol fuchsin three to five minutes. Rinse in water; dip in alcohol. To differentiate and decolorize, place in methylene-green aniline oil from fifteen minutes to two hours, according to the thickness of the section. Ethereal oil; xylol; balsam.

9. Kuehne's Fluorescin and Oil-of-Cloves Method (88). Soak sections in a concentrated aqueous solution of oxalic acid five to ten minutes. Rinse in water: dehydrate in alcohol. Stain in fuchsin-aniline water or methylene blue in ½- to 1-per-cent. aqueous carbonate-of-ammonia solution. Dehydrate in alcohol containing small amount of fuchsin or methylene blue five to ten minutes. Ethereal oil; then xylol; balsam.

10. After Weigert (89). The section is exposed to a saturated solution of gentian or methylene violet in aniline water. If the section is stained on the object-glass, the excess of staining solution is removed by blotting-paper and the iodide-of-kalium solution is dropped upon it. In this case the staining solution is allowed to remain in contact with the section for only a very short time. The sections stained in the saucer are afterward washed in solution of common salt, placed upon the object-glass, dried, and treated with iodide. Afterward again dried with blotting-paper, and aniline oil is dropped upon it several times. The section is then transparent. Remove aniline oil with xylol; then in balsam.

11. Weigert's Modification of Kuehne's (90). Stain in 50 grammes concentrated aqueous solution of violet, to which has been added 1 drop of HCl. Rinse in water; iodide-of-kalium solution; absolute alcohol; pure aniline oil; xylol; balsam.

12. Dry Method, after Kuehne (50). Stain in 1-per-cent. aqueous solution of ammonia

carbonate, to which has been added aqueous solution of methylene blue for ten to fifteen minutes. Rinse in water; decolorize in 1 to 2 per-cent. HCl; wash in water; dry on the object glass by means of a stream of air produced by bellows; xylol; balsam.

Double Stain.

By the use of the common methods in:—

1. Protoplasma Staining. By hardening in chromic or picric acid.

2. Nuclear Staining. With carmine solution, by the common methods, Nos. 4, 6, 7, 8, 10, 11, and No. 6 first in *b* solution washed in water; then several hours in *c* solution.

3 Double Staining of the Decolorized Nuclei:—

(*a*) By Treatment with Alcohol. After first staining in some aqueous solution, rinse in alcohol, then double stain in some contrast color (vesuvin, methlene green); dehydrate in alcohol; clear, and mount in balsam. (May be used with No. 4.)

(*b*) Dehydrate in Aniline Oil. From the xylol the section is placed in aniline oil containing some contrast color (safranin, auramin, eosin, fluorescin) for two to ten minutes, after which the section is transparent; then aniline oil, ethereal oil, xylol, balsam. To be used with Nos. 6, 8, and 9.

Characteristic Staining Methods—Tubercle Bacillus.

1. After Ehrlich (77). Stain section in aniline-water gentian violet or fuchsin for twenty to twenty-four hours. Decolorize in 20- to 30-per-cent. HNO_3 for a few seconds, until only slight traces of the coloring matter is visible; alcohol (70 per cent.) until no more coloring matter is given up; after-stain in methylene-blue or malachite-green or Bismarck-brown solution for a few minutes: rinse in alcohol or water; dry or dehydrate; clear in oil; mount in balsam.

2. After Ziehl-Neelsen (19). Stain section in warmed carbol-fuchsin solution for one hour; decolorize in 5 per-cent. H_2SO_4 for a few seconds; alcohol, 70 per cent.; continue as in Koch-Ehrlich.

3. After Gabett-Ernst (56). Stain the section in cold carbol-fuchsin solution for two to five minutes; decolorize and double stain in methylene-blue sulphuric acid one minute; rinse in water; dry or dehydrate; clear in oil; Canada balsam.

4. After Hermann (52). Stain in a heated mixture of Hermann's solution, *a* or *b*, for one minute; decolorize in $\frac{1}{10}$ nitric-acid four or five hours; wash in alcohol, 95 per cent.; clove-oil, turpentine, xylol, Canada balsam.

5. After Lübimoff (54). Stain for twenty-four hours in cold borax-fuchsin solution; decolorize in H_2SO_4 (1 to 5). Differentiation from lepra bacillus is, that the decolorization does not take place so rapidly.

6. After Kuehne (50):—

(*a*) Stain in carbol-fuchsin ten minutes; decolorize in 30-per-cent. HNO_3; extract in 60-per-cent. alcohol until the section has a rosy color; wash in water; dehydrate in absolute alcohol thirty minutes; place in methylene-green aniline oil, diluted by adding equal parts aniline oil, five to ten minutes; ethereal oil, two minutes; xylol, Canada balsam.

(*b*) Stain in carbol-fuchsin for ten minutes; rinse in water; extract in fluorescin alcohol a few minutes: double stain in methylene-green aniline oil five minutes; ethereal oil, xylol, Canada balsam.

(*c*) Stain in 1-per-cent. aqueous solution of carbonate of ammonium, to which has been added concentrated aqueous solution of crystal violet, for one hour; rinse in water; iodide-of-kalium solution, two to three minutes; concentrated alcoholic solution fluorescin; alcohol, clove-oil, terebene, xylol, balsam.

7. After Kuehne. Triple Staining (50):—

(*a*) Weakly stain in Delafield's hæmatoxylin solution; wash in water several seconds; dehydrate in alcohol; stain in carbol-fuchsin ten minutes; rinse in water; extract fuchsin in fluorescin-alcohol; rinse in pure alcohol; ethereal oil, xylol; place in auramin-aniline oil

until stained a yellowish color; rinse in pure aniline oil; ethereal oil, xylol, Canada balsam. We produce thus a triple staining: nuclei, violet; protoplasm, yellow; bacillus, red.

(*b*) Stain in hæmatoxylin solution; then in a weak solution of extract of logwood containing alum; rinse in water for a long time; absolute alcohol a short time. Stain in a mixture composed of a concentrated alcoholic solution of fuchsin and a solution composed of 1-per-cent. solution of ammonium carbonate, thymol-water, equal parts, for twenty-four hours. Rinse in water; dehydrate in alcohol; aniline oil several minutes; concentrated aniline-oil auramin solution ten to fifteen minutes; turpentine, xylol. The cell-protoplasm is yellowish gray; nuclei, violet; tubercle bacilli, red.

(*c*) Stain the section in nucleus black, diluted three or four times by water, for several minutes, until the section assumes a dark-gray tone. Rinse in a weak aqueous solution of carbonate of lythium until the section is a light-gray color; rinse in water; dehydrate in alcohol for five minutes; stain in carbol-fuchsin ten minutes; wash in water; extract in fluorescin-alcohol; wash in pure alcohol; place in methylene-green aniline oil, not too concentrated, for five to ten minutes: ethereal oil, xylol, balsam.

Bacillus, red; nuclei, vessels, and protoplasm, of different tones of bluish green. The tubercle bacillus is also stained in sections by the common methods of Gram, Kuehne, and Weigert.

Lepra Bacillus.

1. After Unna's Dry Method (91). Stain in aniline-water fuchsin twelve to twenty-four hours. Differentiate in 10- to 20-per-cent. aqueous solution HNO^3 until the section assumes a yellowish color; spiritus dilutus several seconds, until the red color returns; wash out the acid by prolonged rinsing in distilled water, or by dipping once in weak ammonia-water: on the object-glass the excess of water is removed by blotting-paper; careful heating over the flame one to two minutes until the section is absolutely dry; immediately mount in chloroform balsam.

2. After Lutz-Unna's Iodide Pararosaniline Method, Modified from Gram's Method (92). Stain in a heated dilute aniline water gentian-violet solution until the section shows an intense dark-blue violet color; then it is carried from one to the other of the following agents, allowing to each a few minutes,—iodide-of-kalium solution, absolute alcohol containing 10- to 50-per-cent. fuming HNO_3, and then pure absolute alcohol. The latter part of this process may be repeated several times (laterally the iodide solution may be omitted) until the section shows only a bluish-green or slate color; then clear in thymol or oil of cloves; balsam.

3. After Baumgarten (93). Stain in dilute alcoholic or concentrated aqueous solution of fuchsin for twelve to fifteen minutes; decolorize in acidulous alcohol (1 part HNO_3, 10 parts alcohol) one-half minute; wash in water; dehydrate in alcohol, oil, balsam.

Differentiation of the tubercle bacillus, inasmuch as they do not stain at all in so short a time.

4. After Luebimoff (54). Stain the section one-half to twenty-four hours in borax-fuchsin, then place in H_2SO_4 (1 to 5) for a few seconds, until the dark-brown colors change to a yellow-brown; then wash in alcohol, oil, balsam.

The methods of Gram, Kuehne, and the various tubercle stains are all practicable. To differentiate the bacillus of tuberculosis the bacillus of leprosy is not stained by Loeffler's alkaline methylene-blue solution (Neisser).

Syphilis Bacillus.

1. After Lustgarten. Stain in aniline-water gentian violet twelve to twenty-four hours at room-temperature, and afterward at 40° C. for two hours; rinse in absolute alcohol several minutes: place in a 1½-per-cent. aqueous permanganate-of-potassium solution ten seconds; place in an aqueous solution of sulphurous acid (produced by treating metallic copper with H_2SO_4 one or two seconds); rinse in water; repeat the process from treatment with permanganate of potassium until the section appears completely colorless; alcohol, oil of cloves, xylol, Canada balsam.

2. After de Giacomi (81). Stain in hot aniline-water fuchsin twenty-four hours; decolorize by first placing in much diluted and then in concentrated solution of chloride of iron; rinse in alcohol; oil of cloves, xylol, Canada balsam.

3. After Doutrelepont and Schutz (91). Fuchsin in 1-per-cent. aqueous solution of methylene violet twenty four to forty-eight hours; decolorize in dilute HNO_3 (1 to 15) several seconds; alcohol, 60 per cent., five to ten minutes; when a pale-blue color, place in a weak, transparent, aqueous solution of safranin for a few minutes; the intensely red section is then placed in 60-per-cent. alcohol several seconds; rinse in absolute alcohol for just a moment; dehydrate; clear in cedar-oil; balsam.

The bacilli are blue, the nuclei and tissue light-red, and round cells (Ehrlich's cells) are blue with red nuclei.

Bacillus Mallei.

Stain in Loeffler's methyleno blue several minutes. Differentiate in aq. destill., 10 cubic centimetres; conc. H_2SO_4, 2 drops; 5-per-cent. oxalic acid, 1 drop, for five seconds; absolute alcohol; cedar-oil; balsam.

After Kuehne (95). From alcohol to water; stain in carbol methylene blue three to four minutes; acidulous water (500 drops water, 10 drops HCl) for a few seconds until the section assumes a pale-blue tone; wash in distilled water; dehydrate in alcohol for a moment; aniline oil, with the addition of 6 to 8 drops oil of turpentine, 5 minutes; turpentine-oil. xylol; contrast stain in oil of turpentine, containing 2 drops of an aniline-auramin solution, or 5 drops of safranin solution; xylol; Canada balsam.

The bacilli appear blue on a rosy or greenish ground.

Typhus Bacillus.

The best stain is Loeffler's methylene-blue solution or Ziehl's carbol-fuchsin for twenty-four hours; rinse in water; alcohol, cedar-oil, balsam.

May also be stained by Kuehne's carbol methylene-blue solution; by other methods they remain colorless, and are not capable of double staining.

LABORATORY INVENTORY.

Microscope, with Abbe and oil immersion.
Koch's steam-sterilizer, with wire basket, etc.
Thermostat.
Gas-pressure regulator.
Hot-water funnel.
Meat-press.
Ice-chest.
Scales.
Water- and sand- bath.
Gas-burners, with one and three jets (Koch-Pfeil or Griffin).
Leveling apparatus for preparing gelatin plates.
Apparatus for counting colonies.
Stand, containing knife, scissors, pincettes, spatulas, inoculation and other needles.
Platinum needles and öses.
Potato-knife.
Syringes, after Koch, Petre, Stroschein.
Glass bulbs: capacity, ½, 1, 2, and 3 litres.
Erlenmeyer's test-tubes, made of hard glass; very long, for Gurber's anaerobic cultures; also Liborius's.
Saucers, double, for potato-cultures.
Watch-glasses.
Glass bell for damp chamber.
Plates, in copper box.
Pipettes, in copper box.
Glass benches and glass rods.
Funnel.
Glass slides and object-glasses.
Jars for preparations and mice.
Rubber tubing and caps.
Clamps.
Cotton.
Filter-paper, pasters, litmus-paper.

REAGENTS, CHEMICALS, STAINS.

Distilled water.
Alcohol, ether, xylol, chloroform.
Acids: sulphuric, nitric (fuming), acetic, chromic, osmic, oxalic.
Salts: natrium; kalium, lithium, ammonium carbonates; chloride of sodium, hydrate of sodium, potass-ammonium alum, iodide of kalium, chloride of iron, and sulphate of iron.
Iodine, metallic copper.
Thymol, sublimate, carbolic acid
Picric acid, tannin.
Oils: clove, cedar, bergamot, aniline.
Ethereal oils: oil of thyme, oil of terebene.
Paraffin, celloidin, vaselin.
Glycerin.
Canada balsam.
Peptone, cell-gelatin, agar-agar.
Fuchsin.
Gentian violet.
Bismarck brown (vesuvin).
Malachite green, methylene green.
Fluorescin, eosin.
Dahlia, Victoria blue.
Carmine, hæmatoxylin.
Planter's nuclear black, Collin's black.
Safranin.
Auramin.

INDEX.

(The numbers refer to tables.)